Proceedings of the 1998 Imported Fire Ant Research Conference

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DIVISION OF AGRICULTURE

Local Arrangements Committee
Donna Shanklin, Chair
Lynne Thompson, Doug Petty, Angela Cochran

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Proceedings of The 1998 Imported Fire Ant Conference

Table of Contents

A brief overview of the National Fire Ant Strategy for Technology: Development, Testing, and Implementation

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Implementation of the Texas Imported Fire Ant Research and Management Plan

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Background & Current Status. In June 1997, the Texas Legislature approved a 6-year Texas Imported Fire Ant Research and Management Plan, providing $2.5 million for the first two years of the plan, beginning Sept. 1, 1997, for fiscal years 1998 and 1999. To date, 35 directed research and education projects have been funded under the Plan at participating institutions and external competitive grant recipient universities that include: the Texas Agricultural Experiment Station (TAES) and Texas Agricultural Extension Service (TAEX), both within The Texas A&M Univerisity System (TAMUS); Texas Tech University (TTU), The University of Texas (UT); and, the Texas Department of Agriculture (TDA).

Program Structure and Participants. The Director of the Fire Ant Project is Dr. James G. Butler, and Dr. Bastiaan "Bart" M. Drees serves as the Project Coordinator. Three committees influence the activities conducted under the plan: The Fire Ant Research & Management Plan Account Advisory Committee or FARMAAC; Fire Ant Research & Management Plan Initiative Committee or FARMIC; and, the Peer Review Committee.

The Fire Ant Research & Management Plan Account Advisory Committee (FARMAAC) contains members that were appointed by the governor and contains representatives fiom industries and public sectors greatly affected by the fire ant infestation, and administration representatives from participating institutions. This committee oversees the entire project, directs resources and approves all actions taken. Members of the committee for 1998-1999 include:

Bob McCan, Chairman (Agriculture)*
Sharon Johnson (City Manager)*
Robert Werner (Citizen)*
Michael E. Etchinson (Public Utility Commission)
Benny Mathis (Structural Pest Control Board)
Robert Looney (Oil and Gas Association)
Bill Messer (Chemical Association)
Marcus Peterson (Texas Parks and Wildlife Department or TPWD)
John Sneed (Texas Department of Agriculture)
Ray Frisbie (Texas A&M University)
Robert Albin (Texas Tech University)
Larry Gilbert (University of Texas)

(*Governor's appointees)

The Fire Ant Research & Management Plan Initiative Committee (FARMIC) contains entomologists and technical representatives from participating universities and agencies. These twelve individuals provide technical input to FARMAAC, monitor the progress of funded projects and components, and many conduct funded research and education programs themselves. Members for 1998-1999 include:

Ray Frisbie, Chairman (TAMUS)
Dick Auld (TTU)
Larry Gilbert (UT)
John Sneed (TDA)

Jerry Cooke (TPWD)
Frank Gilstrap (TAES)
Bart Drees, Coordinator (TAMUS)
Bradleigh Vinson (TAES)

Harlan Thorvilson (TTU)
Ed Vargo (UT)
Charles Onstad (USDA, ARS)
Lloyd Wendel (USDA, APHIS)

The Peer Review Committee will evaluate the technical merits and progress of funded projects on an annual basis. Their summary report will be used by FARMAAC to consider changes in funding for future years. Members represent specific discipline areas that relate to Project activities. Committee members for 1998-1999 include:

Cliff Lofgren, Chairman (1998)
Sanford Porter (USDA,ARS) - biological control
Chip Taylor (University of Kansas) - genetics & management
Coby Schal (North Carolina State University) - physiology & behavior
Beverly Sparks (University of Georgia) - extension
Robert Taylor (Auburn University) - economics

Funded Research. The plan recognizes three broad areas of research to address the imported fire ant: 1) sustainable solutions; 2) pest management solutions; and, 3) deliverable technologies.

Sustainable solutions include approaches which could conceivably result in sustained suppression of fire ant populations. Discipline areas which fall into this research category include biological control and genetic manipulation. Biological control agents currently under investigation include: Phorid flies (*Pseudacteon*) by Larry Gilbert (UT); *Beuveria bassiana* (fungus) by Harlan Thorvilson (TTU); *Pyemotes* sp. (Acari) by Marilyn Houck (TTU); and, native natural enemies (Protozoa: Microspora (*Thelohania solenopsae*) and Strepsiptera (*Orasema* sp.)) by Bradleigh Vinson (TAES, TAMU). Funded imported fire ant genetics projects are being conducted by Loren Skow, Spencer Johnston, Kevin Heinz (TAES, TAMU).

Pest management solutions include those which might require periodic input of a product, that would also require an investment in time, labor and money. Projects funded are those with the potential of improving currently available Integrated Pest Management (IPM) programs, which rely heavily on cultural and chemical tactics. Several principal investigators are investigating the potential for modifying the fire ant's physiological processes or behavior, including Ed Vargo (UT), Richard Dislippe (TTU), Bradleigh Vinson and Larry Keeley (TAMU). Pest management alternatives for specific usage sites are also being investigated, including: electrical equipment by Harlan Thorvilson (TTU); livestock production systems by Charles Barr (TAEX) and Andy Herring (TTU); and, wildlife by Brad Dabbert (TTU). Novel biological/biochemical approaches and processes are also being investigated, including: research on nutritional enzymes by Edgar Meyer (TAMU); G-protein coupled receptors by Patricia Pietrantonio (TAMU); pheromone and odorant receptors by Robert Renthal (UT San Antonio); and, vibrational and chemical cues by Bradleigh Vinson (TAMU)

Survey and Regulatory Programs. The Texas Department of Agriculture program component supports projects to support regulatory and survey efforts. That agency's survey, quarantine and regulatory programs are under the supervision of David Kostroun and John Sneed. Part of their funding allocation has been designated to support research projects in this area. Robert Coulson

(TAMU) has been designated as coordinator for these survey efforts. He is heavily involved in information delivery systems, particularly in GIS-based applications. Surveys of non-infested counties along the western edge of the distribution of the red imported fire ant in Texas are being conducted by Harlan Thorvilson (TTU) and Ed Vargo (UT). Computer simulation models are under review, and new models, based on existing data, are being developed by Ted Wilson (TAMU). Factors limiting the western spread of the ant are being investigated by Bill Mackay (UT, El Paso).

Educational Programs. The Texas Agricultural Extension Service (TAEX) is the lead agency to conduct educational programs and demonstrate deliverable technologies. The focus of the educational programs will be to promote the concept that fire ants can best be managed in urban areas when conducted on a community-wide basis. The funding provided to implement the Texas Plan has allowed the Extension Service to hire four Extension Agents - IPM (Fire Ants): Scott Russell for Dallas and Tarrant Counties; Lisa Butts for Williamson and Travis and Counties (Austin); Pam Traylor for Harris County (Houston); and, Nathan Riggs for Bexar County (San Antonio). They will be recieving technical support from Charles L. Barr, Extension Associate, Fire Ant Project - IPM Research, who will also be conducting applied research to demonstrate deliverable technologies and improving IPM programs for agricultural commodities.

Economic Evaluations, Program Impact and Public Relations. A number of economists and program evaluation specialists have been funded to conduct economic evaluations and impact assessments of the implemented plan, including: Eduardo Segarra (TTU); Ron Lacewell, John Ellis, Curtis Lard, Charles Hall, Raghaven Srinivasen (TAMUS); and Scott Cummings (TAEX). Media relations will be conducted by Agricultural Communications personnel: Lynette James (Coordinator), Gena Parsons, Edith Chenault, Penny Banks, Blanca Jackson (TAMUS), Jacqui Lockaby (TTU), Ramona Nye (TDA), Robert Meckel (UT), and Tom Harvey (TPWD).

A number of educational products have already been generated to support and promote implementation of the Texas Imported Fire Ant Research and Management Plan. The world wide web site (fireant.tamu.edu) can be accessed to see virtually all of the educational products produced and released to date. A newsletter, *Fire Ant Trails,* has been initiated to keep program participants and colleague up to date about program activities. Fourteen Fire Ant Plan Fact Sheets are under development, and some of these are available as print-on-demand documents and are posted on the web site. Two Extension publications have also been developed to support the plan: B-5043, "Managing Red Imported Fire Ants in Urban Areas"; and, L-2061, "House-infesting Ants and Their Control."

Economic Impact of Texas Fire Ant Plan. In 1995, there were an estimated 17,665,000 people living in Texas (J. Cummings, 1995, *Texas Handbook*. Moon Publications, Inc. 598 pp.). At a cost for implementing the Texas Imported Fire Ant Research and Management Plan of $2.5 million per year, the per person cost of the program is $0.14 per year. Four fifths of these funds are directed towards supporting basic research in biological control, genetics, physiology, economics, survey and integrated pest management (IPM). One fifth of the funds support Extension education programs, which are largely directed towards encouraging the implementation of community-wide fire ant management programs. Thus, the per person cost for fire ant education is $0.03 per year. The Extension Service's targeted county program areas for

Dallas/Tarrant Counties (3,885,400 people), Travis/Williamson Counties (870,000 people), Bexar County (1,302,100 people) and Harris County (3,301,900 people) reach 9,359,400 people, or 53 percent of the population of Texas.

Acknowledgment

All those receiving funding from the Texas Imported Fire Ant Research and Management Plan wish to give special thanks to Representative Tom Ramsay for introducing House Bill 2341 (1995) and his tireless support to seek funding to implement the Texas Plan.

"Research Focus Concerning the Fire Ant at Texas A&M under the Texas IFA Legislative Initiative."

S.B. Vinson

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My goal here is to outline the major "basic" research focus of the new Texas A&M University Imported fire ant program as approved under the Texas Imported Fire Ant Legislative Initiative. The management program and the economic evaluation programs are not covered here.

The overall goal of the research program is to provide fundamental information about fire ants and their natural enemies that can be exploited in their management. The research is focused in three major research areas. These are: a) Sustainable solutions, b) Pest management solutions, c) Deliverable technologies. The sustainable and pest management parts of the research program comprise the "basic" research component. The deliverable technologies part of the research program is focused on evaluating the effectiveness of present management approaches, and to determine the effectiveness and aid in the development of new management approaches as they are discovered. This is a major responsibility of Dr. C. Barr and is not discussed further here.

The Sustainable Solutions research program is concerned with developing fire ant management approaches that are either self sustaining or require a low level of input on a yearly basis. There are four research categories outlined in the plan. These are: 1) Parasites and diseases: 2) Competitors and predators: 3) Genetic manipulation: 4) Environmental constraints. The environmental constraints part of the research is concerned with trying to identify conditions that are unfavorable or favorable to fire ant growth, development and reproduction. If such information were available it might be possible to manipulate the population of ants and to more accurately determine areas at risk of invasions. For example, fire ants appear to not prefer shaded areas. Can shading an area reduce fire ant activity? While these questions are not presently a major focus of the sustainable research efforts at Texas A&M, the survey work described below will collect environmental data that may provide some important clues to some of the environmental constraints.

Of the sustainable solutions research program the major activity and interest has been in the potential use of parasites and diseases. As is evident in this issue of the proceedings, most of this interest is directed towards the importation and release of exotic biological control agents. This classical approach has been very successful in some situations. However, another biological control approach that has potential for fire ant management is augmentation. This involves the use of native parasites and diseases. In the United States there are six native parasites and diseases of the imported fire ant that have been discovered. Their impact has been minimal due to their isolated distribution, low abundance, poor dispersal and ant defense. The Texas A&M program focuses on mass production, development of release technology and augmentation of these organisms which could reverse the above contraints. The Texas A&M Program is working on the three native natural enemies that are starred. This research is divided into two projects using three of the six native natural enemies. The six native natural enemies are: a) Parasitic strepsiptera (Caenocholax) * b) Parasitic worms (Nematodes), c) Parasitic wasps (Orasema)* d) Parasitic fungi (Beauveria)*, e) Protozoan diseases (Thelohania) *, and f) Parasitic mites (Pymotes)*.

The first biological control project has one objective applied to three of the potentially important species. The objective is to determine the incidence, distribution, biology, spread and methods of invasion of fire ant colonies by each of these organisms so that methods to either encourage or promote these organisms can be developed. This project presently involves J. and T. Cook, J. Martin, R. Gold and S.B. Vinson with J. Heraty collaborating with us in our efforts to study Orasema. We are also coordinating our efforts concerning Thelohania with D. Williams of the USDA.

The second biological control project is focused on three objectives. These are: A) Develop *in vitro* mass culture of these biological control organisms. There are three subobjectives for A. These are: 1) Determine composition of hemolymph, 2) Development of IFA cell cultures; and 3) Development of a rearing media. B) Develop release technology for these organisms. C) Develop augmentation procedures. Our initial efforts are directed towards objective 1. Dr. F. Consoli is focused on this project.

Another area of research that is considered "sustainable" is determining the major predators and competitors of fire ants. This research area involves two projects here at Texas A&M. The first project is concerned with the interaction of fire ants with other organisms in regard to competition for resources as well as space. This work is directed towards the impact that the fire ant is having on wildlife and is focused on small mammals were studies can be replicated. Through replicated studies we can collect the quanity of data that is needed in order to determine principals that can be applied to game and domestic animals. Interactions that we are investigating include direct attack, competition for food (insects, seeds) and space, and have the different life strategies of the different species of small mammals influence their risk of an effect. This is a continuation of past research and is a collaborative effort between Entomology (S.B. Vinson) and Wildlife (W. Grant). T. Bedford is also presently involved in this project.

The second predator and competitor project is more concerned with the interactions between fire ants and their ant competitors and predators as they impact the establishment, abundance and distribution of fire ants. The goal is to determine the most effective the fire ant predators and competitors and to develop ways that they can be encouraged. This work is essential to determine where control efforts should be focused and when they should be used. The specific objectives for this project are divided between two approaches, a biological and a management approach. A) <u>Biological</u>. Determine which native ants are good competitors. This research is focused on two sub-projects. These are: a) Impact of select native ants on IFA colony foundation. b) native ant resource- competition. 2) Determine how the more effective competitors compete. 3) Biology of the more effective competitors. 4) Biology of foraging and colony defense in fire ants. B) <u>Management</u>. 1) Response of native ants to fire ant control approaches. 2) Develop fire ant specific baits 3) Develop fire ant specific applications. The work presently involves A. Rao, and J. Martin, but some additional people will be added.

The third Sustainable research area is in Genetics. We have three broad objectives under this research area. These are: A) Clone and characterize the IFA genome B) Develop techniques to characterize and define IFA populations C) Determine the genetic and developmental basis of male sterility. For objective A, we are constructing a genomic library of the IFA as a basic step in providing support to the genetics and molecular program. Once constructed it will allow for the isolation and development of the species-specific molecular markers and allow for the isolation of specific genes of interest including those involved in male sterility. This work is being led by Dr. L. Skow (Genetics) and Dr. S.B. Vinson, (Entomology) along with Dr. S. Johnston (Entomology) and S. Davis (Animal Science). Objective B is concerned with the identification and characterization of fire ant populations in Texas including hybrids. We have developed microsatellites, but other techniques are being developed. The third project is to determine ploidy, male fertility, sperm development and loci responsible for sex determination and fertility. This project is a collaboration between Dr. S. Johnston and Dr. S. Vinson and has four subobjectives. These are: A) Determine the distribution and level of male sterility in the Texas population. B) Define the developmental biology of sterility. C) Determine the physiological basis of stylopization which also leads to male sterility. D) Use microsatellites to map the sex loci.

The second major research area in the Texas Research and Management Plan is termed: Pest Management Solutions. There are 2 major areas that the Texas A&M program has focused on with seven subprojects. The two major areas are physiology and behavior. The following are the subprojects in each. A) Physiology: 1) Food Flow-Nutrition, 2) Digestion, and 3) Reproduction B) Behavior: 1) Communication and 2) Competition. Of the three physiology projects the first concerns the flow of resources into the colony and the environmental factors that control both sex allocation and a reproduction in fire ants. This is being examined by Indira Kuriachan and S.B. Vinson. We are presently examining the environmental factors that initiate worker and gyne production. We are also investigating the factors that influence reproductive dominance in polygyne colonies. The second physiology project concerned with endocrine regulation of queen reproduction focussed on flight muscle aptosis and vitellogenin control which is a collaborative effort between Dr. L.L. Keeley and Dr. Vinson. The objectives: a) Determine the hymolymph factors involved in initiating flight muscle aptosis (breakdown). b) Isolate and identify hymolymph factors involved in reproductive control c) Isolate and characterize the genes involved in vitellogen production. The third physiology project is concerned with a structure-based design of fire ant digestive enzyme inhibitors which is a collaboration between Drs. E. Meyer, S. B. Vinson, and J. Powers (Georgia). The objectives of this project are: a) Isolate and characterize the enzymes involved in protein digestion. b) Design and produce inhibitors to these enzymes, c) Determine the effect of these inhibitors on food flow in IFA colonies.

The second "Pest Management Solutions" research focus area is on behavior and the behavioral modification of fire ants. Communication among colony members is essential to colony survival. Unlocking these codes can open up the opportunity to interfere with colony success. Such communication involves chemical, sound, vibration, visual and contact cues. Most research has been conducted on integrative chemical pheromones. We propose to focus on sexual selective pheromones and nonchemical factors involved in social communication. The major focus is on the role of pheromones and vibrational cues in colony integration and sexual selective factors. The project is being directed by Drs. S. B. Vinson, H.J. Williams, and S. Rauth. The objectives are: A) Determine the role of sound in communication and social integration of fire ant colonies. B) Develop bioassays so that the pheromones involved in initiating mating flights and in sex location can be isolated and synthesized. C) Isolate and identify non or low volatile fire ant repellents.

Another area that involves research, but is not part of the basic research program, is concerned with ant survey's in Texas. These ant surveys are to serve several purposes. One is to aid the Texas Department of Agriculture in surveying boarder counties for the presence or absence of the imported fire ant. This is important to the quarantine responsibilities of the Texas Department of Agriculture. Another survey is concerned with determining the nature and extent of the Imported Fire Ant infestation in Urban areas as part of the fire ant management plan. For the management program to function it is important to know whether the ants are native or imported, as we want to preserve native competitors of the imported fire ant. We also need to know what forms, of the fire ants, polygyne or monogyne, are infesting a particular area so that the proper treatments can be applied. A third survey is directed to the development of a GIS/landscape ecology approach to ant distribution and abundance. The focus will be on the several forms of Imported Fire Ants and the major fire ant competitors. This project is being directed by Dr. R. Coulson, Dr. S. B. Vinson and M. Guzman.

Lastly, there are two other basic research efforts being conducted at Texas A&M. Dr. Pietrantonio is isolating "G-protein-coupled receptors" (GPCR'S) that are surface receptors which mediate numerous critical physiological functions. Dr. Heinz is investigating the possibility of developing a DNA delivery system.

Arkansas' Fire Ant Abatement Programs: Successes and "Learning Experiences"

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http://www.uaex.edu/natural/fireant/firehome.htm

The red imported fire ant (Solenopsis *invicta*) has been present in Arkansas since the late 1950's. Arkansas, like many other fire ant infested states attempted various eradication programs. In the 1990's the focus turned from eradication of fire ants to education of Arkansans. Fire ant abatement programs are a direct result of the education process. Abatement, the reduction in degree or intensity, is presented to Arkansans as a management option to minimize the impact of fire ants. Once people become aware of the history and biology of this pest and that eradication is not possible, they are receptive to working together to impact fire ant presence in their community. Several communities have responded to the concept and have initiated programs to reduce the presence of fire ants.

The following is just a brief overview of several community programs that have been initiated in Arkansas since 1993 and some of the lessons we have learned along the way.

JunctionCity, Arkansas
JunctionCity is a small community of approximately 700 residents in Union County, Arkansas. The program was initiated by American Cyanamid's interest in determining if an aerial application of a bait product (Amdro®) would be feasible. Local officials were presented with the concept and aerial applications were initiated in October 1993. Cooperation among various entities including local civic organization, schools and the Cooperative Extension Service (CES) was important. School children were sent home with notices about the flyover. Evaluation of the effectiveness of the application was also a part of the program. Post-application control was very good and it appeared as though the program could continue since many problems had been overcome; including coverage of non-labeled areas prior to application and fears about human toxicity of the bait product. The city decided NOT to continue the program after the 1994 application due to costs – they couldn't afford the cost of the pesticides and the aerial application fee.

Lessons Learned

- Coordination among different agencies is possible
- Due to the culture of the people changes needed to be made. A majority of homeowners in JunctionCity have spring gardens, making spring aerial applications of Amdro difficult due to label restrictions
- The product can work in reducing fire ant numbers, but the project can fail due to financial constraints of the community involved.

Texarkana, Arkansas

Texarkana, Arkansas (Miller Co.) is a large community of 22,600 residents. The local extension committee in 1993 determined that the fire ant issue was the number one issue on which they wanted Extension to development a program. Under the direction of Doug Petty, a pilot program of approximately 60 houses was initiated in 1993. In 1998, there are approximately 400 household involved in the program. The City of Texarkana and the CES cooperate fully in the program. Using mosquito abatement programs as a model, neighborhood are placed in the program if at least 80% of the people in the neighborhood sign up. Voluntary fees are paid on a square foot basis. The fees are used to purchase the bait material, and pay the salary of the city employee. Block coordinators are an important part of the program. It is their willingness to go out and ask neighbors to join that makes the program work year after year. A similar program is ongoing in Hope, Arkansas (Hempstead Co.) *(See http://www.uaex.edu/natural/fireant/texark.htm for more detailed information)*

Lessons Learned

- Cooperation among agencies is possible
- People can work together for a common goal
- It is hard to retain good employees in an abatement program

Little River County, Arkansas

Little River County is a county of 13,000 residents. A 'reduced-cost' program was initiated by community interest and the local governments desire to do something to help the people. The local government wanted to do something that would meet the county residents needs as well as those living in the city. The County Judge, Clyde Wright, and the quorum court developed the idea of selling bait products at a 'reduced cost'. They would purchase bait material in large enough quantities such to obtain a cheaper price. Baits were selected to be sold due to their effectiveness, and the knowledge that often the price of the material keeps many people from purchasing it. Since 1994 baits have been sold to county residents and it is working very well. The program meets the needs of county residents, and doesn't entail too much work on the part of local government. *(See http://www.uaex.edu/natural/fireant/litriver.htm for more detailed information)*

Lessons Learned

- Rural community needs are different than the urban community
- A "reduced cost" program is effective in getting people to use baits
- A "reduced cost" program is 'easy' to start and requires little more than the initial start up money, a place to store the pesticide, and a day to sell the product

Fordyce, Arkansas

Fordyce is a community of 4,700 residents in Dallas county. A neighborhood abatement program was initiated at the request of Fordyce residents. CES was involved in the sign-up and the city government was responsible for the actual applications. Local officials promoted the program and it was off to a good start with over 60 people signed up. However, today, this program has folded. It folded for several reasons. One of the primarily reasons was the fact that no one person or group, other than CES, took responsibility for the success of the program.

Lessons Learned

- It is hard to retain good employees in an abatement program.
- Total cooperation of all involved people is important for a program to sustain itself.

Cossatot Conservation District, Sevier Co. Arkansas

In 1998, a new program was initiated in Sevier Co., Arkansas (pop 13,700). Brought on by county and city government officials desire to aide the residents of the county with the fire ant problem. A two-pronged approach of selling bait products at a 'reduced cost' and treating neighborhoods, cemeteries, and city parks in the larger communities differentiates this program from others. The mayor of DeQueen, and the County Judge pledged money for the initial purchase of materials, and the local National Resource Conservation Service (NRCS) pledged personnel to be involved in the program. Robin Stacy, NRCS technician, has organized and applied for grants to sustain the program. CES is involved in providing the education support for the program, and advice on proper treatment methods.

Lessons Learned

- A multi-agency project is possible
- There can be more than one approach for a community
- Monies are available through federal and state grants to initiate abatement programs

Overall Lessons Learned

Extension's role in the entire fire ant abatement process has been education. Although we have taken the lead role in helping to initiate or organize abatement programs – we ultimately leave it up to the community to take 'ownership' or responsibility for the program. Extension is still in the process of trying to determine what are the reasons some programs succeed while other fail.

We have learned that each county or city has it own issues with fire ants, and the response varies. Communities that have lived with fire ants for a long time are much more complacent than communities that have been infested within the last 5 - 10 years. As with any program, one person can affect the success of the program. However, cooperation among different groups and individuals is possible. People all have the same goal – to minimize the impact of fire ants on their daily lives.

As with any fire ant program it is clear that an educated CES agent(s) is at the center of fire ant education process of Arkansans. We continue to search for better ways to inform agents in a quick, but accurate format of the management options available to the various segments of the population impacted by fire ants.

Incorporation of Fire Ant Control in the County Extension Program

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Background: Incorporation of fire ant control methods in a county extension program is an important aspect of a county agent's job in a fire ant infested area. Miller County, located in the southwest comer of Arkansas is an ideal location for fire ants due to its location. The Sulfur and Red River valleys consist primarily of dark black and red soils in which the fire ants appear to survive very well. The area also is the most densely populated in this region, allowing the fire ant to be a concern for a greater number of people.

Extension areas: The Cooperative Extension Service has long been involved in helping groups to organize and solve problems. We also are looked upon by businesses and groups as an unbiased source of information. Fire ants affect all areas of life in Miller County and programs are conducted in all four areas of Extension programming: Agriculture, Community Development, Home Economics, and Youth. In each of these areas, the Extension Service has conducted numerous educational efforts, in both public and private enterprises, to reduce the impact of this pest. Efforts include providing educational materials, conducting demonstrations, organizing groups, and conducting meetings. From 1992 through 1997 we have held 122 meetings, 47 demonstrations and 16 TV interviews related to fire ants. (See Table 1)

Agriculture: In the past, fire ant control in agriculture was mainly aimed at control practices for pastures and hay meadows. Today, we address many other areas including: poultry houses, electrical units, baled hay, soybeans, and grain sorghum. Agricultural educational efforts have used local organization's meetings, field days, publications, media, newsletters, displays, individual contacts, and demonstrations. Some of the demonstrations conducted in Miller County include: Fire Ant Control in Pastures, Fire Ant Control in Poultry Houses, Broadcast vs. Mound Treatments, Mechanical vs. Chemical Control, Fire Ant Control at Reduced Rates, Fire Ant Control in Baled Hay Using Anhydrous Ammonia, and Bait vs. Chicken Feed Taste Test.

In an attempt to look at alternative methods to control fire ants for agriculture producers a test using anhydrous ammonia on baled hay was conducted to determine if it had the potential to control fire ants in hay bales. Producers are impacted by fire ants in hay due to the fire ant quarantine which will not allow shipments of hay outside the quarantine unless it has been treated (currently there is not legal treatment of baled hay). Anhydrous ammonia has been used for years to improve the quality of poor quality hay and we thought it might fit into a producer's management system. Twelve square bales of hay were inoculated with 100 ml of fire ants, allowed to sit overnight, then fumigated with 1%, 2%, and 3% anhydrous ammonia. All treatments achieved 100% control. We will continue to look at other management methods.

Community Development: One of the biggest successes of the Miller County Cooperative Extension Service, was the organization and development of the Neighborhood Fire Ant Abatement Program (NFAA). Texarkana's NFAA was one of the first successful organized efforts to control fire ants in residential areas. This program, implemented in 1993, has consistently achieved more than 97% reduction in fire ant numbers and has been the model for other programs in Arkansas and other states (See Table 2).

In developing the NFAA, the first step was to identify the local needs. This was done through a County Extension Council. They prioritized the problems with the fire ant to determine which problem should be given the most emphasis. We then researched the methods of control, and past efforts at community control programs. A plan was then developed to reduce fire ant populations in large areas to a level which they would not cause significant problems. After the plan was developed, the council and local officials had to 'be sold' the program. The initial plan was agreed upon by all those involved. We limited the number of participants initially to be able to manage the program. However, varied sites in different neighborhoods exposed the plan to different conditions. Over the five years of the program we evaluated the program and adjusted the program to address any problems. Today, it has become a self sustaining program in which Extension's role is to advise and monitor. In 1998, there are more than 400 households involved in the NFAA.

Two major industries have received assistance in the development of control programs. A detailed plan was developed, emphasizing proper timing, integrated pest management, and varying types of treatments for a local pulp mill. Through working closely with schools and other public facilities, we have established fire ant control programs for all five elementary school playgrounds, three housing authorities, and three city parks, cemeteries, and golf courses. All of these entities treat annually for fire ants.

Youth: A major part of the Miller County fire ant program involves the education of youth. Young people are naturally curious, and want to know more about fire ants than just what treatments will control fire ants, unlike many adults. Young people are more receptive of programs aimed at the behavior and biology of fire ants, giving them an advantage toward problem solving. A variety of methods, teaching tools, and programs are used with youth.

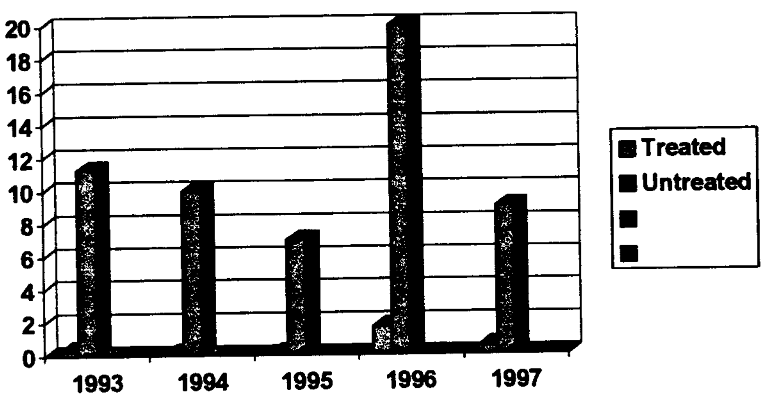
One youth program involved third, fourth, and fifth graders in a study conducted in their school. Fifty percent of the interior of Fairview Elementary School was infested with foraging fire ants. A test was conducted in which only exterior baits were used. After treatment with a pesticide bait, students monitored the impact on the interior foraging of the ants over a three week period. The students were involved in the testing and learned first hand the scientific process used for research.

Conclusion: Educating the public on not only fire ant control measures, but also the fire ant's behavior and biology is needed to develop effective fire ant control programs. Informing people that no one treatment is effective in all situations is part of the education process. We try and teach people that the use of a combination of treatments and cultural practices is required to achieve control of fire ants.

Table 1: Listing of fire ant-oriented extension programs in Miller Co., Arkansas.

Year	Meetings	Demonstrations	Television
1992	21	3	1
1993	19	5	2
1994	30	10	3
1995	17	3	2
1996	26	14	3
1997	15	12	5
TOTAL	**128**	**47**	**16**

Table 2. Yearly results of treated vs. untreated lawns in Texarkana Neighborhood Fire Ant Abatement Program, no. of mounds per 2,500 sq. ft



Effects of Two Experimental Exposures of Turkey Poults to Red Imported Fire Ants

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The red imported fire ant, ***Solenopsis invicta,*** has established itself in North Carolina from the Coastal Plain as far west as Cleveland County and as far north as Guilford, Wake, and Nash Counties, an area that includes much of the major poultry producing regions in the state. In eastern North Carolina where fire ants have been long established, poultry growers have largely accepted their presence. The primary complaint in this region is that foraging fire ants collect on bird carcasses and make it difficult for laborers to remove daily mortality without getting stung. In the western production area (primarily Union and Anson counties) where fire ants are a relatively new pest, more troubling complaints have arisen. Turkey growers have reported young poults scratching the litter away from sidewalls and pecking at ants as they entered turkey houses to forage. Growers claim that the ants are responsible for excess mortality and affect the performance of the poults. Two trials were conducted at N. C. State University to determine whether poults would readily feed on fire ants, and if so, what impact a severe fire ant challenge might have on bird mortality and performance.

Materials and Methods:

Ants for both trials were field collected along Interstate 40 near Warsaw, NC about 70 miles southeast of Raleigh, NC. A teflon coated, 50 ml graduated plastic tube was inserted into large, active mounds. Tubes were left in place for approximately five minutes, retrieved and sealed, placed in a cooler and transported back to Raleigh for use the following day. The total number of ants collected were determined volumetrically. Volume equivalency had been previously determined with exact counts of ten, 20 milliliter samples of freeze killed ants.

Both trials were conducted in an isolation room located in the Poultry Science building on the university's main campus. Trial 1 involved two groups of twenty-five poults, one day of age. Each group was placed in a 4 ft. diameter plastic wading pool containing 2-3 inches of pine shavings. Birds were provided feed and water ad libitum. Supplemental heat was provided by suspending a single heat lamp approximately 18 inches above each pen. Immediately after the birds were placed, approximately 5,000 fire ants were added to pen one. Pen two poults served as unexposed controls. Five birds were removed from each pen for examination at twenty four hour intervals for four days. Birds were weighed, euthanized, checked for external lesions. Birds in the RIFA exposed group were dissected to determine the presence and locations of sting lesions along the digestive tract. Total feed consumption and feed conversion for both groups were calculated after eight days.

Trial 2 was conducted in much the same manner. Fifty, day old poults were allocated equally to one of two treatments. They were penned, fed and watered, and provided with supplemental heat as described in Trial 1. The introduction of fire ants into pen one, however, was delayed until day three of the trial. A smaller number of ants (approximately 3,000) were introduced for this trial. Five birds were removed from each pen for examination on days 4, 5, 6, and 7. All birds and feed were weighed daily.

Results and Discussion:

Exposure group birds in both trials began eating ants as soon as they were exposed. Consumption continued through the first 24 hours, after which birds actively avoided the ants. Birds received numerous stings on their feet and shanks, but no lameness was noted.

Gross examination of birds 24 hours post-exposure revealed stings on the tongue and the lining of the oral cavity, especially in the back of the throat. The ants probably died at this point, as no lesions were noted in the esophagus, crop, proventriculus, or gizzard. Dead ants were present in the crops of all exposed birds. Stings initially appeared as small yellow pustules and by the second or third day post-exposure had ulcerated and crusted over. Many of the lesion had healed by the fourth day post-exposure.

No mortality occurred in either trial. However, birds exposed to fire ants went off feed for approximately three days in both trials and weighed 10% less than controls. Birds in Trial 1 which were exposed to ants at time of placement fell behind controls in body weight within 24 hours (Fig. 1). Birds in Trial 2 were given two days to acclimate themselves to the experimental pens before exposure to ants. They too fell behind in body weight within 24 hours Fig. 2). The period of decreased growth rate in both trials corresponds to the presence of the severe lesions in the oral cavity. Feed conversion values were not different for exposed versus control poults in either study. The difference in body weights was apparently due to decreased feed consumption during the days immediately following exposure to ants.

The ingestion of fire ants and the subsequent decrease in feed consumption has not been documented in the field in North Carolina. It should be noted, however, that the lesions involved are only present for a few days and careful examination within two or three days of ingestion would be required for accurate diagnosis. The short term impact of RIFA stings on poult performance cannot be discounted. Reduced feed intake, even for relatively short periods, compromises early poult performance. Compensatory gain may overcome early growth suppression resulting from lower feed consumption in RIFA challenged poults, but the short time frame of this trial did not address this question. Other basic questions remain unanswered. For example, does stress and the risk of secondary infection directly or indirectly associated with fire ant stings further compromise poult performance? The impact of aversion must also be considered. Since poults clearly avoid fire ants once they had been stung, it is possible that they may continue to refuse feed if large numbers of fire ants are foraging in feeders.

Figure 1: Trial 1 poult body weights (control = square; RIFA exposed = circle).

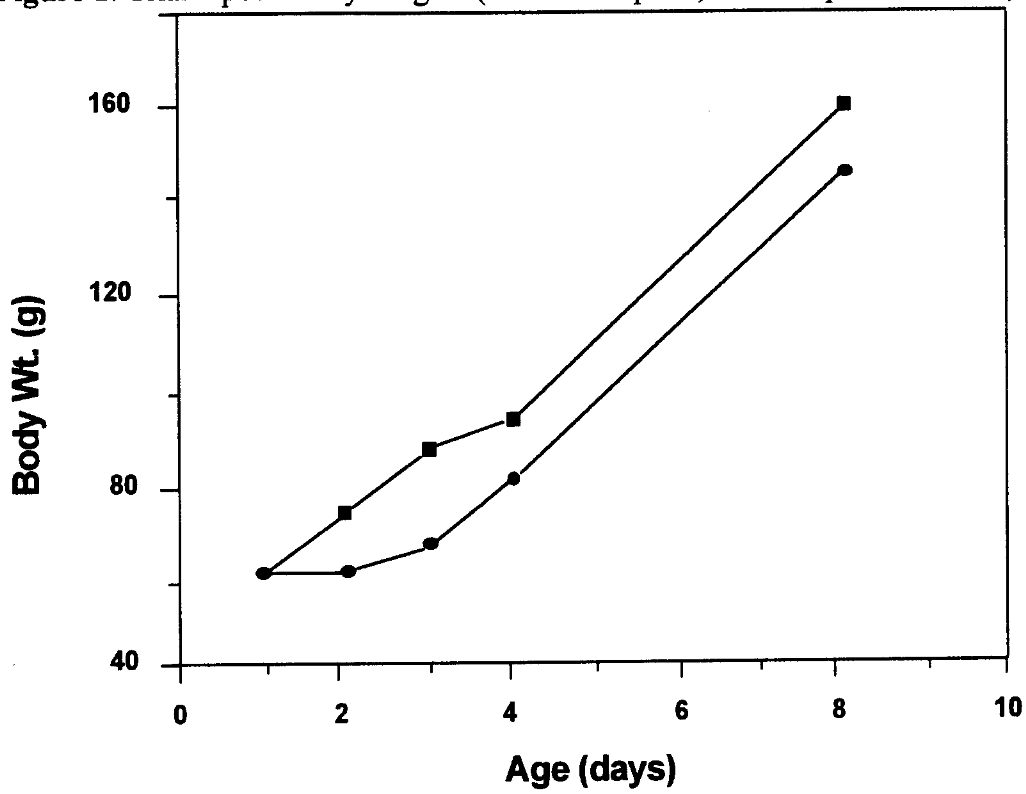
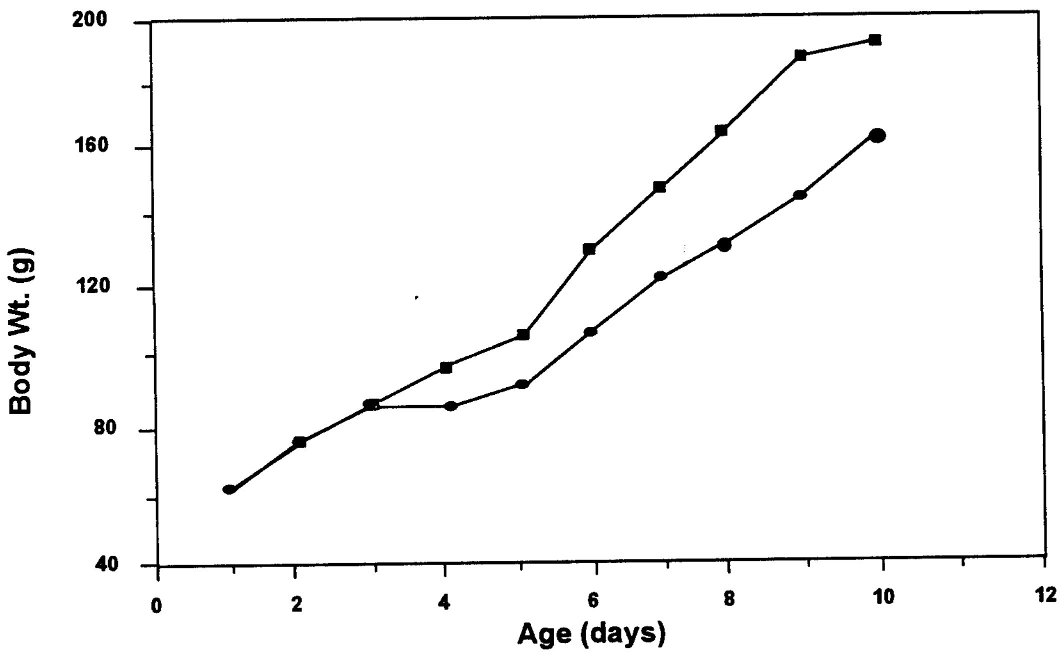


Figure 2: Trial 2 poult body weights (control = square; RIFA exposed = circle).



Induced Red Imported Fire Ant Behavior Towards a Static Electric Device

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From March 1996 through 1997, the Electric Power Research Institute (EPRI) and Texas Tech University's (TTU) Departments of Engineering Technology and of Plant and Soil Science agreed to study methods to protect electrical equipment from infestation by red imported fire ants (RIFA). The basic purpose of the contract was to continue development of an electronic device which exploited certain aspects of RIFA behavior and prevented the RIFA from infesting electrical equipment.

A static electric device (SED) was developed for such a purpose. The basic design consisted of a transformer supplying AC power to metal grid plates where ants were shocked. RIFA were extremely attracted to the SED when workers were shocked and released alarm pheromones. Yet, the working transformer alone (70 VAC) might have had a significant role in fire ant attraction to the SED. The objectives were to determine if: (1) fire ants were more attracted to a working SED transformer as compared to a non-working transformer, (2) an immediate attraction to a working transformer existed, and (3) differences in numbers of ants aggregating under working transformers as compared to non-working transformers existed.

Attraction to Active and Non-active SED3 Transformers

Materials and Methods. One individual tray (55 x 44 x 13cm) was cleaned and disinfected with 70% EtOH. Two SED3s were placed in the tray, one each at opposite ends of the tray lengthwise, and transformers of each SED3 touched the tray floor. A piece of nylon string was used to draw a line 8.0 cm from each device. A brood box filled with ants was placed in the center of the tray. Both SEDs were 17.0 cm from the brood box, and ants had equal access to both devices. After placement into the tray, ants were given 30 minutes to become familiar with the environment, after which one SED3 was turned on. The active SED3 transformer did not supply current to grid plates, which prevented ants from being shocked. The ants then were given 30 minutes to readjust. After which, all ants on the devices or behind the 8.0-cm designated line were removed with an aspirator and placed in separate containers. Removed ants were placed back with the colony after each observational period. Counting started after both observational areas behind designated lines were cleaned. Once counting started, ants that crossed each designated line toward either SED3 were aspirated and placed in a corresponding container. Each observational period lasted ten minutes, and three observational periods for each of the ten replications were completed. After collection, ants were placed into a freezer until frozen. Ant weight and head capsule widths were measured in mg and in mm, respectively.

Student's t-tests were used to determine differences in total ant numbers collected between active and non-active transformers. Also, a t-test was used to determine differences between weight and head capsule widths of ants collected near electrified and non-electrified transformers.

Results. A significant difference was detected for mean total number of ants collected near the electrified and non-electrified devices. Significantly more ants crossed the 8.0-cm line and were attracted to the active SED3 transformer (41.1 ± 12.2) as compared to the inactive SED3 transformer (6.8 ± 6.3) ($t = 3.1$; $df = 1$; $P < 0.008$). Significant differences were not detected for head capsule widths nor for weight of ants that crossed the line toward SED3 transformers. Therefore, no size difference in ants was noted, but more ants were attracted to the SED3 with an active transformer.

Initial Attraction to Active Transformer

Materials and Methods. One individual tray measuring (55cm x 44cm x 13cm) was cleaned and disinfected with 70% ETOH. Two SED3s were placed in the tray, one each on opposite ends of the tray lengthwise. One transformer was active and one was not. A brood box filled with ants was placed in the middle of the tray. In the first series of experiments, both SED3s were placed 15.0 cm from the brood box. In the second series, both SEDs were placed 10.0 cm from the brood box. The transformer of each SED3 touched the tray floor. The ants had equal access to both devices. After placement into the tray, ants were given one minute to become familiar with the environment, after which counting started. All ants crossing the 8.0-cm designated line were counted for one minute. Fifteen replications were conducted for each series of experiments. Each replication consisted of a different colony brood box filled with ants placed into the tray. An analysis of variance was used to determine differences in total numbers collected between active and non-active transformers.

Results. A two-way analysis of variance detected no differences in number of ants moving toward transformers (on/off) ($F=0.56$; $df=59$; $P=0.456$). However, significant differences in number of ants were detected between distances of the SED transformer ($F= 29.95$; $df = 59$; $P<0.001$). No interaction among the variables existed ($F=0.29$; $df=59$; $P=0.594$).

Ant Aggregation Under Transformer

Material and Methods. Twenty different ant colonies in trays measuring (55 x 44 x 13 cm) were each subjected to one SED3. Ten colonies were subjected to one active SED3 transformer, and ten colonies to an un-electrified SED3 transformer. Numbers of ants under SED3 transformers were recorded every 30 minutes for an eight-hour period. Transformers touched the tray floor, and no current was supplied to the grid plates. A paired t-test was used to determine differences among the electrified transformers.

Results. Significantly more ants were counted under electrified transformers of an SED3 (3.7 ± 2.1) as compared to an un-electrified transformer (0.2 ± 0.4) ($t = 21.6$; $df = 169$; $P<0.001$). Ants preferred to aggregate under working transformers used on the SED3.

Discussion

Red imported fire ants were attracted to electrified transformers of a static electric device designed at Texas Tech University. No differences were detected in the size of responding ants. However, distances of transformers from brood boxes were important in ant response to electrified and non-electrified SED transformers. More ants aggregated under electrified transformers in response to undefined stimuli. In the field, ants pile soil and debris near electrical equipment carrying current. These activities cause malfunction, corrosion, and occasional shut-down of electrical equipment that may affect human convenience and safety.

Acknowledgments

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Logic™ - Methods of Application for Fire Ant Control in Pasture and Label Update

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Fipronil: A Molecule for All Reasons

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INTRODUCTION:

Fipronil is a broad spectrum pyrazole insecticide under development by Rhone-Poulenc Ag. Co. The insecticide acts as a GABA blocker in the insect and is active either by ingestion or contact. Fipronil has shown a broad spectrum of activity against many arthropods. Currently, this product is registered in the U.S. is for mole cricket control on golf courses.

Presented here are results from numerous trials from 1993 through the present where we have found excellent activity against imported fire ant (IFA) when granular formulations are incorporated into potting media or applied broadcast to grass sod. Bait formulations have also shown excellent potential against IFA.

MATERIALS AND METHODS:

Containerized Nursery Stock:
Trials have been conducted on site at the Gulfport Plant Protection Station and at several commercial nurseries within the IFA infested area to evaluate the efficacy of granular fipronil as a preplant incorporation treatment. Cooperating nurseries include: Wight Nursery (Cairo, GA), Windmill Nursery (Folsom, LA), Flowerwood Nursery (Mobile, AL), and Turkey Creek Farms (Houston, TX).

In all trials, we blended fipronil granular insecticide (0.1% - EXP 60818A) into potting media provided by each nursery on site or at the Gulfport site. The media contained all fertilizer and nutrients normally used by the nursery. After blending, treated media was potted up in trade gallon nursery pots and aged on site under normal conditions of irrigation and other cultural practices. Media aged at the Gulfport site was subjected to simulated nursery conditions, including supplemental irrigation of 1-1½" per week. Rates of application ranged from 5 to 50 ppm.

At monthly intervals, three pots from each treatment rate were collected, composited, and sent to the our lab. We subjected the treated media to a standard alate female bioassay. Test chambers were 2.5" x 2.5" plastic flower pots which were equipped with a Labstone® bottom. Labstone is generally available through dental supply firms such as Patterson Dental Co. (2323 Edenborn Ave., Metairie, LA). The labstone bottom prevents the queens from escaping through the drain holes in the bottom of the pot and also serves as a wick to absorb moisture from an underlying bed of wet peat moss. Ants are susceptible to desiccation so humidity/moisture levels must be optimized. Plastic petri dishes were inverted over the tops of the pots to prevent escape from the top of the test chambers. Prior to placing queens in the test chamber, 50 cc of treated potting media was placed in the bottom of each pot. Due to possible pesticide contamination, test chambers were discarded after use. Each treatment to be evaluated was subdivided into 4 replicates; with one test chamber per replicate. Five alate queens were then introduced into each replicate. All evaluations were based on a 7 day continuous exposure period, i.e. introduced queens remained in the test chambers for 7 days. At this time the contents of each chamber were expelled into a shallow laboratory pan and closely searched for the presence of live IFA alate queens. Percent mortality was recorded.

Grass Sod/Turfgrass:
Test sites were located on grass sod at Woerner Turf Farm (Elberta, AL) in July 1996, and at municipal airports in Camden, AR; Hattiesburg, MS; and La Porte, TX in May-June 1997. Fipronil 0.1% granular insecticide (EXP 60818A) was applied with a Herd® Granular Applicator mounted on a farm tractor. Liquid treatments (Dursban® 50WP) were applied with a roller pump boom sprayer. The spray rig was equipped with three TKSS tips which provided a 10 ft. swath. The system was operated at 50 psi

providing 37.5 gallons of finished spray per acre. Each treatment was replicated three times. Rates of application, and formulations tested are as follows:

TREATMENT	RATE OF APPLICATION
1996 Fipronil EXP 60818A (0.1G)	12.0 lbs formulated material/acre
1997 Fipronil EXP 60818A (0.1G)	12.5 lbs formulated material/acre
1997 Fipronil EXP 60818A (0.1G)	18.75 lbs formulated material/acre
1997 Fipronil EXP 60818A (0.1G)	25.0 lbs formulated material/acre
Commercial std: 1996 turf trial - Talstar® 0.2G 1997 airport trial - Dursban® 50WP	 200 lbs formulated material/acre 16.0 lbs formulated material/acre
Untreated check	-

All test plots were 1.0 or 0.8 acre in size, with a ¼ acre efficacy subplot located in the center of the test plots. The 1996 turf trial involved effects on IFA populations only. The 1997 airport trials evaluated effects of fipronil on various insect populations: ground pearls at Texas, cutworms at Arkansas, and fire ants at Mississippi. IFA populations were evaluated at all sites as secondary pests. This report will discuss only the IFA evaluations. IFA population counts were made before application and at 6 week intervals thereafter using the population indexing system described by Lofgren and Williams (J. Econ. Entomol. 1982, 75: 798-803). Colony mortality and changes in pretreatment population indices were calculated. Data was statistically analyzed by ANOVA and treatment means separated by LSD test ($P \leq 0.05$) for each posttreatment rating interval.

Bait Trials:
Trials were initiated in June 1996 in Harrison Co., Mississippi, and in May-June 1997 at the above mentioned airport locations. Fipronil bait 0.0015% (EXP 61443A - 15 ppm) was applied to 0.5 acre plots in 1996 due to limited material, and to 0.8 acre plots in 1997 at a rate of 1.5 lbs. material/acre. Amdro® was also applied at 1.5 lbs/acre as a standard at all sites. Bait applications were made with a shop-built granular applicator mounted on a farm tractor. IFA population evaluations were

made as described above.

RESULTS:

Containerized Nursery Stock:
Many trials are ongoing, but initial data indicate that about 15 months of residual activity is provided at 25 ppm and at least 24 months of activity is provided by 50 ppm rate.

Efficacy of Fipronil 0.1% Granular (EXP 60818A) when Incorporated in Various Potting Media.

Rate (ppm)	Months Residual Activity						
	Flowerwood		Windmill		Wight	Turkey Creek	MAFES
5	1	--	0	--	9	--	--
10	4	6+	2	0	13	15	6+
15	--	6+	--	0	--	15	6+
20	--	6+	--	--	--	15	6+
25	15+	6+	18	4	13	15	6+
35	--	--	--	9+	--	--	--
40	--	6+	--	5+	--	--	6+
50	15+	6+	28+	5+	31+	--	6+

Grass Sod/Turfgrass:
Twenty-eight weeks of excellent control was provided in the 1996 turf trial (Tables 1 & 2). Unfortunately, the plots were harvested after that evaluation so that the actual longevity of the treatment could not be determined.

The 1997 airport trials are still being evaluated, but results to date will be presented. The Camden, AR site was heavily infested with imported fire ants. Initial colony counts at this site averaged 204 colonies/acre, indicating that this may be a polygynous site. Exhumation and examination of numerous nests indicated that over half of the colonies examined had multiple de-alated females with worker ants congregating around them. Therefore, this site was considered polygynous.

Granular formulations of fipronil at all sites have provided excellent control of IFA for a minimum of 18 weeks after treatment, providing 92-100% reduction in colony numbers and 95-100% reduction in population indices throughout the period (Tables 3-8). While there were no statistical differences in the various rates of fipronil at any site, efficacy of the 12.5 lb/acre rate in Texas dropped to 75% control at 24 weeks after treatment. The two higher rates at Texas and all rates at the Arkansas site have continued to provide 98-100% control (both reduction in colony numbers and population indices) through 24 weeks. The Mississippi site has maintained 91-100% control of IFA at all rates through 40 weeks. Control with fipronil was similar to or slightly better than the Dursban standard at all sites and all evaluation times.

Bait Trials:
In both the 1996 and 1997 trials, there was no significant difference at any evaluation period between the fipronil bait treatment or the standard Amdro treatment (Tables 9-16). Fipronil provided >83% control of IFA through 18 weeks at all sites. Reinfestation was noted at most sites by the 24 week evaluation.

CONCLUSIONS:

Fipronil has a number of possible use patterns for control of imported fire ants. As a preplant incorporation treatment, granular fipronil (0.1%) provides ca. 15 months of residual activity at 25 ppm and more than 24 months at 50 ppm. Future trials with a 0.05% formulation may provide more consistent results at ≤25 ppm.

In grass sod/turfgrass, fipronil 0.1% granular (EXP 60818A), at rates of 0.0125, 0.01875 and 0.025 lbs ai/acre, effectively controls IFA in turfgrass. Test plots will continue to be monitored to determine length of residual activity provided by the higher rates of application.

Trials this spring will evaluate a 0.05% granular formulation at the same ai rates of application.

Fipronil formulated as a 15 ppm bait on defatted corn grit carrier (EXP 61443A) provided control equivalent to the Amdro standard. This spring trials will be initiated evaluating a 0.00015% (1.5 ppm) applied at 15 lbs material/acre. This will provide the same amount of ai per acre, but the higher rate of application may make application easier, especially for homeowners.

Rhone Poulenc Ag. Co. will be pursuing EPA registration for both the granular and the bait formulations of fipronil in the next year.

Mention of trade names or proprietary products does not constitute an endorsement or recommendation by the U.S. Department of Agriculture.

Table 1. Efficacy of Candidate Grass Sod Treatments - reduction in pretreatment population index - 1996.

Treatment	Rate of Applic. (lb material/ acre)	Mean % change in population index at indicated wks PT			
		(6)	(12)	(20)	(28)
Fipronil EXP60818A	12.0	-100a	-100a	-82.3a	-100a
Talstar flowable	2270 ml/acre	-99.2a	-100a	-100a	-100a
Talstar granular	200.0	-95.1a	-39.5a	78.2ab	--
starch suSCon	40.0	-77.4a	-87.7a	-74.9a	--
Untreated check	----	-39.3b	85.3b	207.6b	-37.5b

Means within a column followed by the same letter are not significantly different according to a LSD test (P=0.05).

Field harvested prior to 36 week count.

Table 2. Efficacy of Candidate Grass Sod Treatments - colony kill - 1996.

Treatment	Rate of Applic. (lb material/ acre)	Mean % decrease in number of colonies at indicated wks PT			
		(6)	(12)	(20)	(28)
Fipronil EXP60818A	12.0	100a	100a	83.3ab	100a
Talstar flowable	2270 ml/acre	95.2a	100a	100a	100a
Talstar granular	200.0	90.5a	52.4bc	0.0c	--
starch suSCon	40.0	77.3a	85.9ab	66.4b	--
Untreated check	----	32.1b	12.1c	0.0c	21.2b

Means within a column followed by the same letter are not significantly different according to a LSD test (P=0.05).

Field harvested prior to 36 week count.

Table 3. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Decrease in Number of Colonies; Camden, AR. Airport, 1997.

Treatment	Rate of Applic. (lb/ acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*			
			(6)	(12)	(18)	(24)
EXP60818A 0.1G	12.5	232.0	98.9a	99.5a	98.2a	100.0a
EXP60818A 0.1G	18.75	132.0	96.1a	100.0a	92.4a	100.0a
EXP60818A 0.1G	25.0	181.3	99.2a	100.0a	97.0a	98.4a
Dursban	16.0	222.7	100.0a	100.0a	91.5a	88.5b
Check	--	142.7	53.5b	85.6b	61.8b	20.5c

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 4. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Change in Population Index; Camden, AR. Airport, 1997.

Treatment	Rate of Applic. (lb/ acre)	Mean Pop. Index Pretreat (per acre)	% change in population index at indicated weeks PT*			
			(6)	(12)	(18)	(24)
EXP60818A 0.1G	12.5	3257.3	-99.7a	-99.9a	-99.3a	-100.0a
EXP60818A 0.1G	18.75	1638.7	-98.2a	-100.0a	-95.4a	-100.0a
EXP60818A 0.1G	25.0	2297.3	-99.9a	-100.0a	-98.3a	-99.6a
Dursban	16.0	3100.0	-100.0a	-100.0a	-92.9a	-92.3b
Check	--	1873.3	-58.1b	-88.2b	-66.3b	-32.0c

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 5. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Decrease in Number of Colonies; Hattiesburg, MS. Airport, 1997.

Treatment	Rate of Applic. (lb/ acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP60818A 0.1G	12.5	26.7	95.8a	95.2a	100.0a	95.8a	95.8a
EXP60818A 0.1G	18.75	30.7	100.0a	100.0a	100.0a	100.0a	100.0a
EXP60818A 0.1G	25.0	32.0	100.0a	95.8a	95.8a	91.7a	95.8a
Dursban	16.0	29.3	100.0a	100.0a	100.0a	100.0a	100.0a
Check	--	29.3	3.3b	33.3b	25.6b	31.1b	17.8b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 6. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Change in Population Index; Hattiesburg, MS. Airport, 1997.

Treatment	Rate of Applic. (lb/ acre)	Mean Pop. Index Pretreat (acre)	% change in population index at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP60818A 0.1G	12.5	360.0	-99.4a	-99.3a	-100.0a	-97.0a	-99.4a
EXP60818A 0.1G	18.75	413.3	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a
EXP60818A 0.1G	25.0	426.7	-100.0a	-99.4a	-99.0a	-94.2a	-97.1a
Dursban	16.0	413.3	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a
Check	--	380.0	43.7b	-46.1b	-34.8b	-15.8b	-14.5b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 7. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Decrease in Number of Colonies; La Porte, TX. Airport, 1997.

Treatment	Rate of Applic. (lb/ acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP60818A 0.1G	12.5	34.7	93.3a	100.0a	100.0a	73.3a	89.2a
EXP60818A 0.1G	18.75	34.7	100.0a	100.0a	100.0a	100.0a	100.0a
EXP60818A 0.1G	25.0	37.3	100.0a	100.0a	100.0a	100.0a	100.0a
Dursban	16.0	48.0	100.0a	97.0a	89.8a	62.1a	91.6a
Check	--	37.3	53.3b	37.1b	4.8b	0.0b	0.0b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 8. Control of Imported Fire Ants in Turfgrass with Fipronil 0.1% Granular - Change in Population Index; La Porte, TX. Airport, 1997.

Treatment	Rate of Appli (lb/ acre)	Mean Pop. Index Pretreat (per acre)	% change in population index at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP60818A 0.1G	12.5	586.7	-99.1a	-100.0a	-100.0a	-75.6a	-91.3a
EXP60818A 0.1G	18.75	522.7	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a
EXP60818A 0.1G	25.0	452.0	-100.0a	-100.0a	-100.0a	-100.0a	-100.0a
Dursban	16.0	889.3	-100.0a	-99.5a	-91.6b	-73.9a	-93.4a
Check	--	513.3	-51.0b	-42.3b	+9.3c	+189.6b	+88.9b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 9. 1996 Bait Trials - population index change.

Treatment	Rate of Applic. lb/acre	% change in population indices at indicated wks PT*			
		(6)	(12)	(18)	(24)
Fipronil	1.5	-96.6a	-96.9a	-94.2a	-67.1a
Fipronil	3.0	-97.3a	-96.2a	-81.7a	-62.8a
Amdro	1.5	-90.1a	-88.3a	-82.4a	-76.0a
Untreated Check	---	-36.3b	-28.2b	12.0b	54.4b

* means within a column followed by the same letter are not significantly different (Tukey's test, P=0.05)

Table 10. 1996 Bait Trials - decrease in number of colonies.

Treatment	Rate of Applic. lb/acre	% decrease in no. of colonies at indicated wks PT*			
		(6)	(12)	(18)	(24)
Fipronil	1.5	81.9a	87.5a	87.4a	61.6a
Fipronil	3.0	84.1a	84.6a	68.6a	56.9a
Amdro	1.5	77.4ab	79.3a	79.6a	76.1a
Untreated Check	---	47.5b	36.3b	3.6b	0.0b

* means within a column followed by the same letter are not significantly different (Tukey's test, P=0.05)

Table 11. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Decrease in Number of Colonies; Camden, AR. Airport, 1997.

Treatment	Rate of Application (lb/acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*			
			(6)	(12)	(18)	(24)
EXP61443A bait	1.5	314.7	97.3a	99.3a	96.7a	92.4a
Amdro	1.5	202.7	90.3a	94.7a	77.2b	74.2a
Check	--	142.7	53.5b	85.6a	61.8b	20.5b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 12. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Change in Population Index; Camden, AR. Airport, 1997.

Treatment	Rate of Application (lb/acre)	Mean Pop. Index Pretreat (per acre)	% change in population index at indicated weeks PT*			
			(6)	(12)	(18)	(24)
EXP61443A bait	1.5	4142.7	-98.8a	-99.9a	-96.9a	-94.9a
Amdro	1.5	2900.0	-90.6a	-96.3a	-80.1b	-79.4a
Check	--	1873.3	-58.1b	-88.2a	-66.3b	-32.0b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 13. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Decrease in Number of Colonies; Hattiesburg, MS. Airport, 1997.

Treatment	Rate of Applic. (lb/acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP61443A bait	1.5	24.0	88.9a	83.3a	94.4a	50.0a	61.3ab
Amdro	1.5	33.3	100.0a	100.0a	100.0a	92.6a	84.3a
Check	--	29.3	3.3b	33.3b	25.6b	31.1a	17.8b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 14. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Change in Population Index; Hattiesburg, MS. Airport, 1997.

Treatment	Rate of Applic. (lb/acre)	Mean Pop. Index Pretreat (per acre)	% change in population index at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP61443A bait	1.5	340.0	-97.5a	-94.2a	-98.8a	-25.8a	-88.6a
Amdro	1.5	453.3	-100.0a	-100.0a	-100.0a	-94.7a	-93.2a
Check	--	380.0	43.7b	-46.1b	-34.8b	-15.8a	-14.5b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 15. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Decrease in Number of Colonies; La Porte, TX. Airport, 1997.

Treatment	Rate of Application (lb/acre)	Mean No. Colonies Pretreat (per acre)	% decrease in number of colonies at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP61443A bait	1.5	42.7	86.9a	90.5a	93.3a	68.5a	91.1a
Amdro	1.5	40.0	90.8a	100.0a	97.9a	58.8ab	70.6a
Check	--	37.3	53.3b	37.1b	4.8b	0.0b	0.0b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Table 16. Control of Imported Fire Ants in Turfgrass with Fipronil 0.0015% Bait - Change in Population Index; La Porte, TX. Airport, 1997.

Treatment	Rate of Applic (lb/acre)	Mean Pop. Index Pretreat (per acre)	% change in population index at indicated weeks PT*				
			(6)	(12)	(18)	(24)	(40)
EXP61443A bait	1.5	702.7	-96.5a	-97.7a	-93.3a	-70.1a	-89.8a
Amdro	1.5	693.3	-98.4a	-100.0a	-98.7a	+9.7ab	-70.4a
Check	--	513.3	-51.0b	-42.3b	+9.3b	+189.6b	+88.9b

* means within a column followed by the same letter are not significantly different, LSD test, P=0.05.

Distance™ - A New Fire Ant Bait for Nurseries and Sod Farms

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Valent USA Corporation
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Spinosad, A Promising New Material for Control of Red Imported Fire Ants

Raymond B. Cooper and T. Craig Blewett
Dow AgroSciences
Indianapolis, Indiana

Spinosad

The first turf and ornamental products in the new spinosyn chemical class of insect pest control materials

Conserve SC is the first DowElanco turf and ornamental insect pest control product in the new spinosyn chemical class.

The new spinosyn chemical class of products combine the positive features of both synthetic chemical insecticides and biological insect pest control products.

The word spinosyn comes from the name of the factors that make up the active ingredient spinosad. Spinosad consists of a combination of factors spinosyn A +spinosyn D.

Spinosad:
Overview

- **Activity: Insect pest control in turfgrass, ornamentals, agricultural and horticultural crops**
- **Active ingredient:**
  Spinosad (pronounced spin-OH-sid)
- **Chemical Class: Spinosyn**
- **Products:**
  - **Conserve* - Turf and ornamentals (greenhouses)**
  - **Tracer* - Cotton/row crops**
  - **Success* - Fruit trees**
  - **Spin Tor* - Horticultural crops**

*Trademark of Dow AgroSciences

Conserve has activity on economically important insect pests in the turf and ornamental markets.

Conserve contains the active ingredient spinosad (pronounced spin-OH-sid) which is in the new spinosyn class.

Conserve is formulated as a 1 lb ai/gallon suspension concentrate and is packaged in a 1 gallon container.

Spinosad: Attributes

- **The active ingredient is derived through the fermentation of a naturally occurring organism (*Saccharopolyspora spinosa*)**
- **Highly active at very low use rates**
- **Combines the efficacy of synthetic insecticides with the environmental characteristics generally associated with biological insect pest control products**

The active ingredient in Conserve is derived through the fermentation of a naturally occurring organism.

Conserve is highly active at very low use rates.

Conserve combines the efficacy of synthetic insecticides with the environmental characteristics generally associated with biological insect pest control products.

Spinosad: Active Ingredient History

- **1982: Soil sample taken**
- **1982-1986: Activity characterization of fermentation broths and soil organisms**
- **1986-1988: New species, <u>Saccharopolyspora spinosa</u>, an actinomycete bacteria identified as source of active fermentation metabolites**
- **1988: Made first sample of this novel fermentation derived compound**
- **1989: Spinosad structure determined**

The history of discovery of the active ingredient in Conserve goes back to 1982.

In 1982, a routine soil sample was taken as part of a collection effort to obtain microorganisms for screening for biological activity.

From 1982-1986, the organisms in the soil sample were grown in fermentation broths and characterization for biological activity took place.

During 1986-1988, an actinomycete bacteria was identified as the source for some of the biologically active fermentation broth derived metabolites. This new species of bacteria was named <u>Saccharopolyspora</u> <u>spinosa</u>.

In 1988, the first sample of the biologically active metabolite was made and in 1989 the chemical structure of the metabolite was determined.

Spinosad:
Active Ingredient Characteristics

- **Crystalline solid**
- **Very low odor**
- **Non-volatile**
- **Water solubility**
  - **Spinosyn A = 235 ppm @ pH 7**
  - **Spinosyn D = 0.33 ppm @ pH 7**
- **Soil sorption moderately strong**
- **Degradation rapid in the environment primarily through photolysis (sunlight)**

The active ingredient in Conserve is a crystalline solid that can be formulated into a suspension concentrate.

The active ingredient also has a very low odor and end-use dilution's of Conserve are practically odor free.

The active ingredient is practically non-volatile and solubility in water is higher for the metabolite spinosyn A than spinosyn D.

The active ingredient has a moderately strong affinity to bind to soil and its persistence in the environment is short due to rapid degradation through photolysis (sunlight).

Spinosad: Safety and the Environment

- **Aquatic:**
  Based on the environmental and toxicological properties of spinosad, there is no need to apply special mitigation measures for uses around aquatic environments
- **Avian:**
  Based upon its LC_{50}, spinosad has been classified by E.P.A. as "Practically Non-toxic" to avian species

Pertaining to Conserve safety and the environment:

For aquatic organisms, based on the environmental and toxicological properties of the active ingredient, there is no need to apply special mitigation measures for uses of Conserve around aquatic environments.

For birds, based upon the LC_{50}, the active ingredient in Conserve has been classified by the E.P.A. as "Practically Non-toxic" to avian species.

Spinosad: Safety and the Environment

- **Based on favorable review of environmental and health data, the EPA placed spinosad under expedited review for registration as a “Reduced Risk” insect pest control material**

Based on favorable review of all of the available environmental and health data, the EPA has placed Conserve under expedited review for registration as a “Reduced Risk” insect pest control product.

Spinosad:
Acute Mammalian Toxicology

- **Rat oral LD_{50}: = 3783 mg/kg (Male)**
  > 5000 mg/kg (Female)
- **Mouse oral LD_{50}: > 2000 mg/kg**
- **Rabbit dermal LD_{50}: > 5000 mg/kg**
- **Rat inhalation LD_{50}: = 5.18 mg/L air (4 hrs)**
- **Rabbit skin irritation: Negative**
- **Guinea pig sensitization: Negative**
- **Rabbit eye irritation: Slight (gone by 2 days)**
- **Formulations: Anticipate a "CAUTION" signal word and a 4 hour WPS reentry interval**

The acute mammalian toxicity of the active ingredient in Conserve indicates that the oral LD_{50} is greater than at least 2000 mg/kg and is greater than at most 5000 mg/kg depending on the test organism.

The dermal LD_{50} is greater than 5000 mg/kg and inhalation toxicity is low.

Skin irritation and sensitization studies were negative.

Test organism eye exposure studies resulted in slight eye irritation with symptoms disappearing within 2 days.

This acute mammalian toxicity profile supports an anticipated "CAUTION" signal word for Conserve and a 4 hour WPS reentry interval.

Spinosad:
Chronic Mammalian Toxicology

- ◆Non-carcinogenic
- ◆Non-mutagenic
- ◆Non-teratogenic
- ◆Non-neurotoxic

Data from studies on the chronic mammalian toxicity of the active ingredient in Conserve indicates that the product is not carcinogenic, mutagenic, teratogenic, or neurotoxic.

Spinosad: Mode of Action

- **MOA distinct from all other insect pest control products**
- **Active on the insect nervous system nicotinic acetylcholine receptors by a mechanism unique among known insect pest control compounds**
- **Active by ingestion and contact exposure**
- **No cross resistance exhibited with other insect pest control products**
- **Because of the unique mode of action, spinosad has a good fit in IPM programs as a resistance breaker**

The mode of action of Conserve is unique and distinct from all other insect pest control products commercially used.

The active ingredient in Conserve is active on the insect nervous system nicotinic acetylcholine receptors by a mechanism unique among known insect pest control compounds.

Conserve is active on the insect nervous system with ingestion resulting in 5-10 times more activity than contact exposure alone.

Cross-resistance studies with insects resistant to other modes of action support the uniqueness of the active ingredient since no cross-resistance has been found. Because of this, Conserve

Spinosad: Symptoms in Insect Pests

- Tremors
- Incoordination
- Paralysis
- No recovery
- Death

Insect pests exposed to Conserve experience symptoms in a specific order.

Upon exposure, the insect pest exhibits tremors that lead to incoordination followed by paralysis. There is no recovery from the paralyzed state and eventually death occurs.

Spinosad: Speed of Activity

- In general, symptoms will begin within minutes after exposure to spinosad with control evident within 1-3 days after treatment
- Feeding stops as soon as symptoms of exposure occur

In general, for insect pests, the symptoms of Conserve will begin within minutes after exposure, with control evident within 1-3 days after treatment.

Feeding stops as soon as symptoms of exposure occur.

Spinosad:
Effects on Beneficials

- **Spinosad has less impact on certain predatory beneficial insects than most synthetic insect pest control products**

–Minute pirate bug:	**LC_{50} = 200 ppm**
–Ladybird beetle:	**LC_{50} > 200 ppm**
–Green lacewing:	**LC_{50} > 200 ppm**
–Predatory mite:	**LC_{50} > 200 ppm**
–Earthworm:	**NOEC > 970 mg/kg**
–Whitefly parasitoid wasp:	**LC_{50} = 29.1 ppm**
–Honey bee: Highly active	**LC_{50} = 11.5 ppm**

The maximum labeled rate for Conserve for insect pests on ornamentals is 200 ppm.

Testing to determine the toxicity of the active ingredient in Conserve to beneficials indicates LC_{50} values greater than or equal to 200 ppm for the predators: minute pirate bugs, ladybird beetles, green lacewings, and predatory mites. This means that Conserve has less impact on certain predatory beneficial insects than most synthetic insect pest control products.

In general, toxicity is low for earthworms, moderate for most parasitic wasps, and is high for honeybees. Avoid application during times when the plants which are intended to be treated are also being frequented by honeybees.

Spinosad: Pest Spectrum

- Coleoptera (Chrysomelid leaf beetles)
- Diptera (leafminers, pod gall midges)
- Hymenoptera (sawflies, fire ants)
- Lepidoptera (caterpillars)
- Siphonaptera (fleas)
- Thysanoptera (thrips)
- Acari (spider mites)

Conserve has activity on a broad range of pests. Tests have shown activity against seven arthropod orders. These include pests like Chrysomelid leaf feeding beetles such as elm and willow leaf beetles; serpentine leafminers; pod gall midges; sawflies; defoliating caterpillars such as bagworm, fall webworm, eastern tent caterpillar, gypsy moth, sod webworm, cutworm, armyworm, and others; fleas; thrips and some spider mites.

Conserve has not shown activity on white grubs or sucking insects, such as aphids and chinch bugs.

Spinosad: T & O Insect Spectrum

- Sod Webworm
- Black cutworm
- Fall armyworm
- Fall webworm
- Fire ants
- Fleas
- Elm leaf beetle
- Spider mites
- Gypsy moth
- Azalea caterpillar
- Yellownecked caterpillar
- Bagworm
- Eastern tent caterpillar
- Leafminers
- Thrips
- European sawfly

Spinosad: Summary

- First T&O product in the new spinosyn class
- Derived from a naturally occurring organism
- Highly effective against many insect pests
- Active by ingestion and contact exposure
- Novel mode of action
- No cross resistance = IPM fit
- Very favorable toxicological and environmental profile ("Reduced Risk" registration)
- Little or no impact on most predatory beneficials
- Not shown to be phytotoxic

In summary:

Conserve is the first DowElanco turf and ornamental insect pest control product in the new spinosyn class.

The active ingredient in Conserve is derived from the fermentation of a naturally occurring organism and it exhibits both ingestion and contact activity, but is most active via ingestion.

Because of the unique mode of action, Conserve will provide a good fit in IPM programs as a resistance breaker.

Based on favorable review of all of the available environmental and health data, the EPA has placed Conserve under expedited review for registration as a "Reduced Risk" insect pest control product.

Even though Conserve has little or no impact on most predatory beneficial insects, it is highly effective against many turf and ornamental insect pests.

Conserve has not been shown to be phytotoxic to turfgrasses or ornamentals.

Field Applications of *Beauveria bassiana* Alginate Pellets

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INTRODUCTION

The lack of biological enemies of the red imported fire ant (RIFA) has promoted the dispersal of the pest. Microbes make up the majority of the natural enemies of Solenopsis invicta in South America. Few of these enemies are present in the United States. In Texas, even fewer of these microbial natural enemies exist (Beckham 1980). Low populations of RIFA microbial natural enemies in the U.S. will enable the spread of RIFA to continue. Two fungi, *Conidiobolus* (probably *macrosporus*) and *Metarhizium anisopliae*, were detected in 0.3% and 0.5%, respectively, of nearly 1000 RIFA founding queens collected in College Station, Texas. In laboratory bioassay, M. *anisopliae* caused 100% mortality of 15 RIFA queens after 5 days (Sanchez-Pena 1992).

The black imported fire ant (Solenopsis richteri Forel) is susceptible to Beauveria *bassiana* (Hyphomycetes: Moniliales) in both adult and larval stages. B. *bassiana* was fed to S. *richteri*, and a mortality rate of 90% was observed (Broome 1974). *Beauveria bassiana* causes effective mortality of the RIFA. By introducing infected ants to uninfected ants, Stimac et al. (1990) observed that the pathogen could be passed from ant to ant.

By exposing RIFA's to dried mycelia of Beauveria *bassiana*, Sanchez-Pena (1992) found an effective method of infecting RIFA and causing mortality. When mycelia were introduced in soil, mortality caused by B. *bassiana* was significantly less than in sterile soil. Encapsulation of mycelia in alginate pellets improved the mortality rate of RIFAs caused by the fungus (White 1995). Encapsulation allowed the fungus to survive better in soil, perhaps allowing successful competition against other microbes present in soil.

Beauveria *bassiana* (ARSEF#2484), BB2484, was originally isolated from workers of the Mexican leaf-cutting ant, Atta *mexicana*, collected on the Pacific Plains of Sinaloa, Mexico, in 1986 (Sanchez-Pena, 1992). The BB2484 was re-isolated several times from infected Solenopsis invicta and grown and stored on Sabouraud's dextrose agar medium with 1% yeast extract (SDAY) in the fire ant laboratory at Texas Tech University.

MATERIALS AND METHODS

Broadcast Treatments

This study was performed in Morris County, Texas. For the broadcast application, a 3x3 Latin square was designed to incorporate three different treatments. Each treatment grid was 2500 square feet in size. Prior to treatment, on 20 June 1997, all mounds were activity-rated, marked with flags, and numbered for future reference.

Three separate treatments were applied to grids: *B. bassiana* mycelia encapsulated in alginate matrix pellets (1.0 mm) that were either coated with peanut oil or were not (2 ml peanut oil per 2 g pellets). Control plots were not treated with alginate pellets.

Mounds in these plots were rated four times in the next 13 weeks. At the end of the trial (19 September 1997), mounds were dissected, and locations of pellets and dead ants were recorded. Flags were pulled up at the end of the experiment leaving the Latin square marking stakes for future collection of data.

Fire ants collected in bait cups placed at the edge of the visible mounds on 9 July and 20 July 1997 were brought back to the laboratory . For surface sterilization, ants were placed them in 10% Clorox for 10 seconds and flushed with water. They were then placed in petri dishes on sterile filter paper and observed. Ants, collected from mounds treated with fungal pellets coated with peanut oil, produced fungi that was identified as *Beauveria bassiana* when conidiophores and conidia developed. *Beauveria bassiana* was re-isolated and grown on Sabouraud's dextrose agar and identified again.

Individual Mound Treatments

Fungal pellets coated with peanut oil, fungal pellets with no peanut oil, and control (no pellets) were used to treat individual mounds. In Cass County, Texas, forty-five mounds with ratings of 25 (Harlan et al. 1981, Lofgren and Williams 1982) were located, recorded, and marked with flags prior to treatment. Twenty pellets were spread on the ground around the perimeter of each randomly selected mound. Fifteen of the mounds were treated with the peanut oil-coated pellets, fifteen were treated with the "no oil" pellets, and the remaining mounds acted as controls.

Fire ants collected in bait cups placed at the edge of the visible mounds on 20 July and 11 August 1997 were brought back to the laboratory . Ants were placed in 10% Clorox for 10 seconds and flushed with water. They were then placed in petri dishes on sterile filter paper and observed. Ants, collected from mounds treated with fungal pellets coated with peanut oil, produced fungi that were identified as *Beauveria bassiana* when conidiophores and conidia developed.

was not supported by the broadcast application trial.

Table 2. Mean Mound Ratings from Alginate Individual Mound Application, 9 July - 19 September, 1997. Bextine Property, Cass Co., Texas.

	Mean Mound Ratings[a]			
	Days Post-Treatment			
Treatment[b]	0	8	32	51
Peanut Oil	25.0a	11.0a	9.0a	1.6a
No Peanut Oil	25.0a	24.3b	20.7b	9.7b
Control	25.0a	19.3b	19.3b	17.0c

[a] Means followed by the same letter within a column are not significantly different. (ANOVA, Randomized Design, LSD, $P<0.05$)

[b] Alginate pellets with *Beauveria bassiana* coated with peanut oil, alginate pellets with *Beauveria bassiana* with no oil treatment, or no pellets applied.

From fire ants collected near mounds treated with fungal pellets coated with peanut oil, *B. bassiana* was re-isolated. Ants collected near the control mounds and the mounds treated with pellets without the peanut oil coating produced no *B. bassiana*. This was an indication that treatments with alginate pellets coated with peanut oil attributed to the decline of the mean mound ratings.

ACKNOWLEDGMENTS

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RESULTS AND DISCUSSION

Broadcast Treatments

No significant differences were observed at pre-treatment in activity ratings (Table 1). After 19 days, the mound ratings in the peanut oil-coated pellet treatment grid were significantly lower then the other two treatments. This trend continued throughout the trial. Peanut oil-coated fungal pellets caused significant mortality in the RIFA population.

Table 1. Mean Mound Ratings from Alginate Broadcast Application, 20 June - 19 September, 1997. Morris Farms, Morris Co., Texas.

	Mean Mound Ratings[a]				
	Days Post-Treatment				
Treatment[b]	0	19	30	52	91
Peanut Oil	21.2a	14.0a	9.0a	7.2a	5.0a
No Oil	21.1a	19.6b	20.5b	20.7b	15.7b
Control	20.9a	20.3b	21.7b	18.4b	18.4b

[a] Means followed by the same letter within a column are not significantly different. (ANOVA, Latin Square, LSD, $P<0.05$)

[b] Alginate pellets with *Beauveria bassiana* coated with peanut oil, alginate pellets with no oil treatment, or no pellets applied.

The growth of *B. bassiana* from fire ants treated with fungal pellets was an indication that the decline in mean mound rating was attributed to the application of *B. bassiana* alginate pellets coated with peanut oil. No ants collected near mounds treated with fungal pellets without peanut oil nor the control developed *B. bassiana* infections.

Individual Mound Treatments

Significant differences were observed eight days after treatment (Table 2). The significantly lower mound rating for the peanut oil-coated fungal pellets at both eight days and at 32 days provides evidence that *Beauveria bassiana* caused significant mortality to the RIFA mounds when baited with peanut oil. Interestingly, at 51 days, significant differences were observed among all treatments. This suggests that, given time, fungal pellets that are not coated with peanut oil may cause mortality in fire ant mounds. This

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Tracking Alginate Pellets Using Ultraviolet-reflective Dyes

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INTRODUCTION

Beauveria bassiana may be incorporated in an alginate matrix, exposed to fire ants, and cause ant mortality (White 1995). Effective delivery of alginate-fungal pellets to imported fire ant colonies is necessary for this technology to be valuable in management of fire ant populations. Recently, we have coated alginate pellets with peanut oil and have determined that foraging fire ants readily retrieve pellets and move them to colonies. Furthermore, ***B. bassiana*** mycelia activate from peanut oil-treated pellets and produce conidia that infect fire ants.

Various ultraviolet-reflective, water-soluble dyes may be added to the alginate and mycelial mixtures before pellets are produced. A bright orange dye was chosen from several candidate dyes because pellets could be easily seen under laboratory lights, daylight, and at night under ultraviolet lamps (365 nm, 6-volt; Spectronics Corp., Westbury, NY). Using this tracking technology, pellets can be easily followed in laboratory and field trials to study transport of pellets into brood boxes and mounds by foraging ants. The goal of the studies reported herein was to determine the efficiency of delivery of baited, alginate pellets to fire ant colonies.

MATERIALS AND METHODS

<u>Laboratory Trials</u>

The goal of this trial was to determine if a peanut oil coating of fungal pellets affected the rate at which fire ants pick up the pellets. Treatments were fungal pellets coated with peanut oil and fungal pellets without peanut oil. Six laboratory colonies of similar size and activity in separate trays (55 x 44 x 13cm) with a brood box (clear plastic box, 12 x 12 x 3.5cm, with plaster filling the bottom one-third) in the center of each tray were used in the experiment. Three replications of each treatment were completed.

In each tray, 20 pellets were randomly dispersed, and a dot of yellow, ultraviolet-reflective paint was placed next to each pellet as a position marker. Observations were made in the dark using a ultraviolet lamp. The position of each pellet was noted after 5 min. periods for 1.5 h, and the final location of each pellet was recorded. The amount of time necessary for the ants to transport pellets into brood boxes was recorded.

Field Trials

At the time of application, the fire ants located the peanut oil-coated pellets almost immediately. After 30 minutes, 63.3% of the peanut oil-coated fungal pellets were moved into foraging tunnels by fire ants (Table 2). After two hours, 98.3% of the peanut oil-coated pellets were moved by the fire ants into foraging tunnels. The lack of movement by fire ants of the fungal pellets not coated with oil contributes to the hypothesis that the oil was an attractant.

Table 2. Movement of *Beauveria bassiana* Alginate Pellets by RIFA Foragers into Foraging Tunnels in Field Applications on 9 July, 1997. Bextine Property, Cass, Co., Texas.

	Mean Percentage of Pellets Taken into Foraging Tunnels[b]				
	Minutes After Application				
Treatment[a]	0	30	60	90	120
Peanut Oil	0a	63.3a	70.0a	95.0a	98.3a
No Oil	0a	0b	0b	0b	0b

[a] Fungal pellets coated with peanut oil or fungal pellets without peanut oil
[b] Same letter within time periods are not significantly different (paired t-test, df=2, $P<0.05$)

DISCUSSION

The ultraviolet dye incorporated into alginate pellets allowed observations to be made both in the day and at night. Ants did not appear repelled by the brightly colored pellets and readily retrieved them for colony use. This technology is a useful technique for the study of red imported fire ant foraging in both the laboratory and field experiments.

ACKNOWLEDGMENTS

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Field Trials

To trace the movement of the alginate pellets in the field, 20 pellets were randomly distributed at the perimeter of three mounds of each treatment (alginate pellets coated with peanut oil and alginate pellets with no peanut oil coating). Each pellet was marked with a toothpick painted with yellow, ultraviolet-reflective paint. The treatments were applied at dusk, thus, at nightfall UV lights were used to observe pellet movement by the fire ants. The ultraviolet lights were held 1.0 m above the ground. The number of pellets that were moved into foraging tunnels was recorded.

RESULTS

Laboratory Trials

The fungal pellets that were coated with peanut oil were moved into brood boxes remarkably fast by the fire ants (Table 1). In one hour, the fire ants had moved almost all of the peanut oil-coated fungal pellets into the brood box, and the pellets with no oil were not even touched. The fire ants were very attracted to the peanut oil, and they took the pellets back to the brood chamber where the fungus could activate and infect the colony. However, the pellets did not hydrate and produce conidia in this trial. Further laboratory trials are needed to further explore the effects of *B. bassiana* in this situation.

Table 1. Movement of *Beauveria bassiana* Alginate Pellets by RIFA into Brood Boxes of Laboratory Colonies.

	Mean Percentage of Pellets in Brood Box[b]						
	Minutes After Application						
Treatment[a]	0	15	30	45	60	75	90
Peanut Oil	0a	18.3a	66.7a	86.7a	96.7a	98.3a	98.3a
No Oil	0a	0a	0b	0b	0b	0b	0b

[a] Fungal pellets coated with peanut oil or fungal pellets without peanut oil coating

[b] Same letter within time periods are not significantly different (paired t-test, df=2, $P<0.05$)

Frequent Insecticide Use for Imported Fire Ant Control

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Introduction

Red imported fire ants, ***Solenopsis invicta*** Buren (Hymenoptera: Formicidae), are an important problem in the southern United States. They cause medical and economic hardships and are just a plain nuisance, so many residents try to control them (Thompson and Jones 1994). This has resulted in the development of many different control methods with varying degrees of effectiveness and safety. Although many different insecticides have been tried, they can be narrowed down into two general categories; insecticide baits, such as hydramethylnon and fenoxycarb, and contact insecticides, such as acephate and chlorpyrifos.

A bait is a liquid food attractant mixed with a soluble pesticide incorporated into a granular carrier (Lofgren et al. 1975). Typically baits are slow acting and are applied so that worker ants can find and carry it back to the colony where it is fed to other colony members. This enables it to claim more ants than just those who touch it. However, because most of the bait is usually moved into colonies within 24 hours (Ferguson et al. 1996). fire ants from outside treated areas can quickly re-invade bait-treated territory (Collins et al. 1992).

Contact pesticides, so called because the insect must come in contact with the chemical for it to work, have varying degrees of residual activity against fire ants (Collins and Callcott 1995). Broadcast applications of contact insecticides deliver the chemical to surfaces likely to be contacted by foraging fire ant workers. Controlling foraging workers should eventually eliminate colonies as food becomes unavailable and the ants in the colonies begin to cannibalize their brood (Lofgren et al. 1975). Contact insecticides should also kill new queens colonizing treated areas. The longer an insecticide remains efficacious against fire ants, the longer the time interval between insecticide applications.

Both types of insecticides reduce the number of fire ants. There are situations, however, where there is no acceptable level of infestation. Just ask a parent who wants to let their young child play outside in their yard, but can't because of the threat of fire ant stings. Ideally. when one treats for fire ants the insecticide would control them for an extended period. It is important, however to not create a zone so toxic that it becomes dangerous to humans. What is needed is a treatment that is not overly dangerous to people and animals, yet remains deadly to fire ants for an extended period.

Through personal contact with local residents, it was brought to our attention that atrazine (a herbicide) may have some promise in controlling fire ants. This would be a good situation, since it is a common compound used for weed control in both sod and farming situations. If atrazine is not effective by itself, perhaps when allied with some of the common fire ant insecticides, a longer lasting and ultimately more effective control could be achieved.

This paper reports on frequent applications of several pesticides for possible longer term control of fire ants.

Materials and Methods

Two municipal airport sites in southeastern Arkansas were selected for the study: Monticello and Warren. Each site was divided into four and two square blocks, respectively, measuring 128 m on a side (about 1.6 ha each). Individual blocks were subdivided into four square plots, 64 m on a side (about 0.4 ha each). Each block was located so that a 35 m or wider buffer separated adjacent blocks and all squares had at least two sides exposed to external fire ant infestations (fig.1). The Monticello airport was home to 4 blocks, while the other two were at the Warren airport.

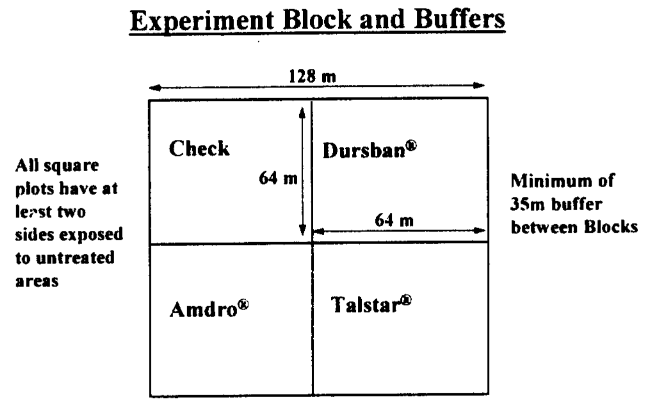

Fig 1. Layout of block with treatment plots.

Atrazine (Atrazine 4L, Sostram Corporation, Roswell, GA), was applied to half the blocks in March 1997, and the other half were untreated. Atrazine is a selective triazine herbicide used to control broad-leaf and grassy weeds in a number of agricultural crops and in turf applications.

Applications of atrazine were made with a portable five nozzle spray rig, mounted to a Polaris 350 All Terrain Vehicle. The herbicide was applied as uniformly as possible by first driving north and south on 3 m wide swaths at 8 km per hour and then finishing by driving east and west once again on 3 m swaths. A mixture of 1920 ml of Atrazine 4L diluted with 115 liters of water was applied to each treated 0.4 ha plot.

Insecticide treatments (bifenthrin, chlorpyrifos, hydramethylnon, and an untreated check) were randomly assigned to the four plots within each block and were first applied in May, and then reapplied every other month (July and September of 1997). Liquid insecticide applications were accomplished using the same spray rig used for the herbicide applications, but cleaned and recalibrated for each compound.

Bifenthrin (Talstar® F, FMC Corporation, Philadelphia, PA), is a pyrethroid insecticide/miticide used in lawn and ornamental applications. Bifenthrin treated plots received 1200 ml of Talstar® flowable per 0.4 ha.

Chlorpyrifos (Dursban® 4L, DowElanco, Indianapolis IN), is a common broad spectrum organophospate insecticide. Treated plots received 960 ml of chlorpyrifos per 0.4 ha.

Hydramethylnon (Amdro® fire ant bait, American Cyanamid, Wayne, NJ), was broadcast over the designated plots using a Herd Model GT-77 Bait Spreader mounted on the back of the same Polaris ATV. The spreader was calibrated to deliver 1.7 kg of bait per hectare when driven 13 km per hour across the area on swaths spaced 7 m apart.

Circular subplots, each 0.1 ha in size, were established at the center of each plot. This center was marked with red flagging and a herbicide (Pramitol® 5G) was applied to a radius of 30 cm to aid future location of subplot centers. A survey of the number of visible fire ant mounds was made on each subplot prior to treatment. A minimum of three people following a 18.1 m rope to define the circular subplot, located and flagged each fire ant mound. Suspect mounds were then probed with a stiff wire to verify the presence of fire ants and were recorded. Visits were made to both sites and subplot mound counts taken prior to insecticide application and repeated at 1, 2, 3, 4 and 5 months after the initial insecticide application.

Statistical significance was determined using ANOVA for atrazine and its interactions and ONE-WAY ANOVA and Tukey's studentized range test for mean separation of insecticides (SPSS Inc. 1997). Data were transformed for analysis using the square root of mound counts.

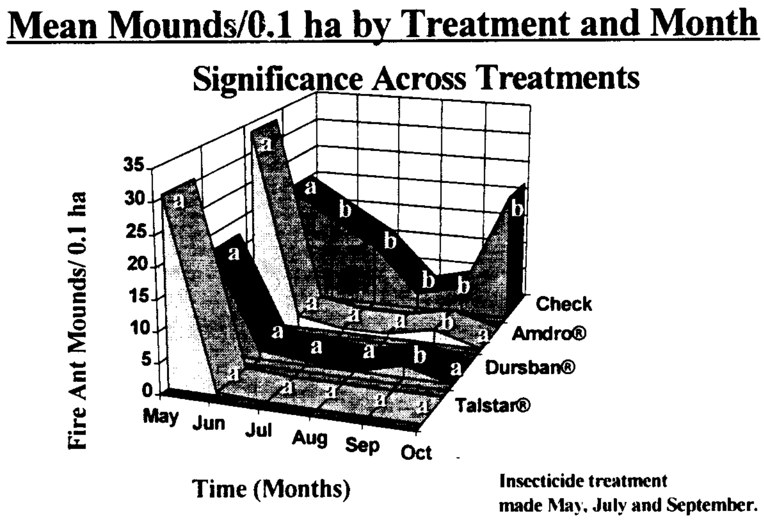

Fig. 2 Imported fire ant mounds per 0.1 ha showing significance across treatments. Means with the same letter within a month are not significantly different at the 0.05 level, Tukey's Studentized Range Test

Results

Data analysis showed that there was no significant difference between atrazine treated blocks and non-atrazine treated blocks ($F_{1, 88} = 0.250$, $P = 0.619$). The atrazine by insecticide interaction was also not significant ($F_{3, 88} = 4.753$, $P = 0.761$). Because atrazine had no effect, the three atrazine treated blocks were combined with the three non-atrazine treated blocks to produce 6 replicates.

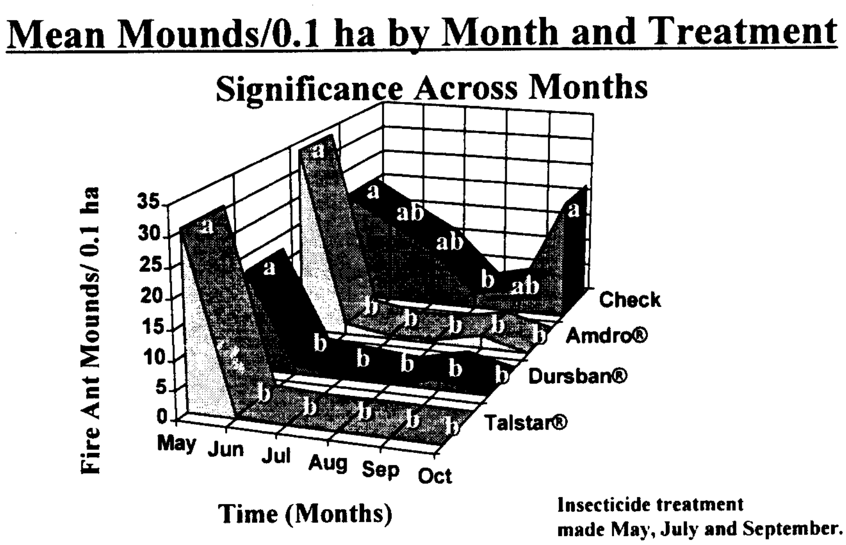

Fig. 3 Imported fire ant mounds per 0.1 ha showing significance across months. Means with the same letter within a treatment are not significantly different at the 0.05 level, Tukey's Studentized Range Test

At the one month post-application date, hydramethylnon, bifenthrin and chlorpyrifos each demonstrated very effective fire ant killing capabilities ($F_{3,20} = 10.7$, $P < 0.05$). All reduced visible fire ant colonies to near zero levels, both across insecticide treatments within months (fig. 2) and across months within insecticide treatments (fig. 3). Repeated applications of the pesticides maintained similar control throughout the course of the experiment. The untreated check plots showed the expected decline in fire ant mounds that is common during the hot, dry summer months, but fire ant populations rebounded to pre-treatment levels by the October counts.

Discussion

Repeated applications at 2 month intervals of all three insecticides (hydramethylnon, chlorpyrifos, and bifenthrin) were effective at keeping visible red imported fire ant colonies at very low levels. Because the multiple treatments were so successful, the frequency of insecticide applications probably could be reduced and still maintain a high level of control.

Acknowledgments

Our thanks to: The City of Warren, Arkansas and the Monticello Airport Commission, who graciously provided land for the experiment; FMC Corp who supplied the Talstar® insecticide; American Cyanamid Co. for providing Amdro® insecticide. This research was funded by USDA, APHIS Grant 97-810-0229-GR, and this report does not necessarily express APHIS's views.

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Measuring Insecticide Efficacy: Counting Fire Ant Mounds Vs. Bait Station Sampling

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Introduction

The primary goal that an investigator works for, when testing imported fire ant, ***Solenopsis invicta*** **Buren** (Hymenoptera: Formicidae), insecticides, is just how effective the compound is at killing fire ants. The methods used to determine efficacy must be accepted as accurate and then withstand tests of peer review. Counting active fire ant mounds in a 0.1 ha circular subplot, along with using population indexing (Harlan et al. 1981, Lofgren and Williams 1982) has been generally accepted as accurate enough to make claims about an insecticide's efficacy.

In previous experiments we noticed that during hot and dry months, fire ant mounds will frequently "disappear" and then "reappear" later when temperatures have dropped and moisture levels are back to normal. Sometimes colonies move for no apparent reason (Hays et al. 1982). This situation hinders research, because it can make efficacy appear better or worse than it actually is. This prompted us to question colony counting to assess efficacy. Perhaps a more accurate method would be to bait the test area, collect the ants that gather on the baits, identify them, and measure efficacy using these data. The absence of foragers would likely mean the absence of colonies.

Many times we have heard homeowners say that the fire ant control product they used caused mounds to move or even disappear, but they still had fire ants! What they were saying was that the absence of visible mounds wasn't as important as the absence of stinging fire ants. Fire ants are very aggressive foragers and are adept at finding available food sources (Ferguson et al. 1996). By nature fire ants are territorial and tend to monopolize a foraging area using their recruiting behavior and ability to utilize trail pheromones (Wilson 1962). Foraging occurs regularly during the life of a fire ant colony except during the founding stage (Markin et al. 1972) and when ambient temperatures are so high or low that worker ants stay home (Markin et al. 1974, Porter and Tschinkel 1987). It seems rational that if food is offered in an area and there are fire ants present, they will eventually find the food.

This study compares two methods for assessing insecticide efficacy. The first is counting mounds (colonies) in a 0.1ha circular subplot and making comparisons over time. This method is fast and easy to accomplish, frequently taking less than 15 minutes per subplot to perform. The downside of this method is that accuracy can suffer because of the fire ant's habit of moving mounds, and not always building visible mounds.

The second method establishes a grid of bait stations over the test area, and collecting all the ants present at the stations after one hour. Ants are identified, counted and comparisons are made over time. We think that accuracy will be much improved because we are sampling ants from large and small colonies. Also, fire ant dominance in the baited area can be mapped. **A** drawback to this method is its time consumption and the fact that training is needed to identify the ants collected.

Materials and Methods

Two municipal airport sites in southeastern Arkansas were selected for the study: Monticello and Warren. The sites were selected for their large size, they are not grazed, and they are mowed frequently making fire ant colonies (mounds) easier to locate. Each site was divided into two or four square blocks measuring 128 m on a side (about 1.6 ha each). Individual blocks were subdivided into four square plots, 64 m on a side (about 0.4 ha each). Each block was located such that a 35 m or wider buffer separated adjacent blocks so that all plots had at least two sides exposed to external fire ant infestations (fig. 1). The Monticello airport was home to blocks 1 to 4. Blocks 5 and 6 were at the Warren airport.

Pesticides

The herbicide atrazine (Atrazine 4L, Sostram Corporation, Roswell, GA) was applied once to blocks 2, 3 and 6 in March of 1997. Blocks 1, 4 and 5 did not receive herbicide treatment. Atrazine was used because we had anecdotal evidence that its use might help control fire ants.

Applications of atrazine were made with a portable five-nozzle spray-rig mounted to a Polaris 350 All Terrain Vehicle. Application of the herbicide was performed by first driving north and south on 3-m wide swaths and then finishing by driving east and west once again on 3-m swaths. Atrazine 4L was applied at the rate of 1.920 liters per 0.4 ha.

Insecticide treatments (bifenthrin, chlorpyrifos, hydramethylnon, and an untreated check) were randomly assigned to the four plots within each block (fig. 2) and were first applied in May, and then reapplied in July and finally in October 1997. Liquid insecticide applications were accomplished using the same spray rig used for the herbicide applications, but cleaned and recalibrated for each compound.

Bifenthrin (Talstar® F, FMC Corporation, Philadelphia, PA), is a pyrethroid insecticide/miticide used in lawn and ornamental applications. Bifenthrin treated plots received 1200 ml of Talstar® flowable per 0.4 ha.

Chlorpyrifos (Dursban® 4L, DowElanco, Indianapolis IN), is a common broad spectrum organophospate insecticide. Treated plots received 960 ml of chlorpyrifos per 0.4 ha.

Hydramethylnon (Amdro® fire ant bait, American Cyanamid, Wayne, NJ), was broadcast over the designated plots using a Herd Model GT-77 Bait Spreader mounted on the back of the same Polaris ATV. The spreader was calibrated to deliver 1.7 kg of bait per hectare when driven 13 km per hour across the area on swaths spaced 7 m apart.

Efficacy

Efficacy was measured using two methods. The first was mound (large colony) sampling. This method created circular subplots, each 0.1 ha in size, at the center of each plot. This center was marked with red flagging and a herbicide (Pramitol® 5G) was applied to a radius of 30 cm to aid future location of subplot centers. A survey of the number of fire ant mounds was made on each subplot prior to treatment. A minimum of three people following a 18.1 m rope to define the circular subplot, located and flagged each fire ant mound. Suspect mounds were then probed with a wire to verify the presence of fire ants. Visits were made to each site and subplot counts taken prior to insecticide application and repeated at 1, 3 and 5 months after the initial insecticide application.

The second method used sugar-bait stations placed approximately 9 m apart in a square-grid pattern such that each treatment plot possessed 36 bait stations (fig. 2). Each bait station consisted of a small piece of cotton dental roll soaked in a 10% sugar-water solution placed in the middle of a white plastic vial lid. Though fire ants are generally considered oil-loving creatures, carbohydrates are essential ingredients in their diet (Vander Meer et al. 1995). For this reason and because we have used sugar water solutions successfully in the past, we used it to bait the fire ants. Bait stations were placed flat on the ground and allowed to attract ants for a minimum of one hour. Ants were collected by snapping a labeled plastic vial over the ants on the lid at each bait station. Vials were brought back to the laboratory and placed overnight in a freezer to immobilize the ants. The next day ants were fixed in 80% alcohol, identified and counted. This procedure was performed prior to insecticide application and repeated at 1, 3, and 5 months after the initial insecticide application.

Mean separations of colony mortality were determined using ONE-WAY ANOVA and Tukey's studentized range test (SPSS Inc. 1997). Data were transformed for analysis using the square root of mound counts.

Baiting results were analyzed using three different methods; 1) mean number of fire ants per bait, 2) percentage of baits occupied by 10 or more fire ants, and 3) the percentage of baits occupied by fire ants. Mean separations were again determined using ONE-WAY ANOVA and Tukey's studentized range test (SPSS Inc. 1997). Count data were transformed using square roots; the arcsine transformation was used for percentage data.

Because mound and bait-station counts measure different characteristics of the population, they are not directly comparable. So, to make comparison easier, the measurements were standardized by creating a ratio using pretreatment counts as the denominator and post treatment counts as the numerator.

Results

Analysis of both mound counting and sugar-baiting showed that there was no significant difference between atrazine treated blocks and non-atrazine treated blocks. Because atrazine had no effect, all the blocks were combined to produce 6 insecticide replicates.

Mound counting

At the one month post-application date, hydramethylnon, bifenthrin and chlorpyrifos each demonstrated very effective fire ant killing capabilities (fig. 3). All insecticides reduced visible fire ant colonies to near zero levels. Repeated applications of the pesticides maintained similar control throughout the course of the experiment (fig. 4). The untreated check plots showed the decline in fire ant mounds that is common during the hot, dry summer months, but fire ant populations rebounded to pre-treatment levels by the October counts.

Bait-station sampling

Collectively all three methods of interpreting the data (mean number of fire ants per bait [fig. 5], percentage of baits occupied [fig. 6], and percentage of baits occupied by 10 or more fire ants [fig. 7]) showed similar results. Hydramethylnon treated plots showed little change the month following initial treatment, and is followed by a slow decline until October when the levels of fire ants finally drop to near zero. The bifenthrin and chlorpyrifos treated plots looked similar, with a precipitous drop in June and continued declines until October when both were near zero. The untreated checks showed a relatively steady level of foraging fire ants, even through the hot, dry summer months. The three methods of viewing the bait-station data showed that there are many foraging fire ants present on the check, hydramethylnon and chlorpyrifos plots. The bifenthrin-treated plots caught almost no foraging fire ants.

Discussion

Comparing mound counts with bait stations gave quite different pictures of insecticide efficacy (figs 8-10). Mound counting samples only the population of visible colonies and in untreated areas large colonies are likely to predominant and thus to be counted. Additionally, ineffective insecticides are likely to kill few ants and thus keep colonies large. Colonies started in May typically grow rapidly and become visible within 3-4 months (Lofgren et al. 1975). Bait-stations sample foraging ants, providing a better representation of ant populations, especially when colonies are small and less visible, which is more likely to occur after insecticide applications and soon after reinvasion of treated areas.

Hydramethylnon is an example of an insecticide that is highly effective in killing colonies, but has no residual activity, so new colonies can be initiated within a few days of its application. Using a computer simulation model, Thompson et al. (1997) showed that fire ants can quickly reestablish on bait-treated sites. They showed that within four months of early summer bait treatments, the number of small fire ant colonies reestablished on a 1-ha site could exceed 2,000 colonies, which is 50 times the pretreatment population of 40 large colonies, and these 2,000 small colonies would produce upwards of 10 million ants, almost 55% of the pretreatment population of 18 million. So, without residual activity to prevent reinvasion, bait insecticides are likely to be ineffective without frequent applications.

Insecticides with residual activity should provide improved fire ant control because they will be killing foraging workers and new founding queens entering from adjacent untreated areas. Our study shows that bifenthrin is somewhat better than chlorpyrifos in keeping fire ants at bay. Because chlorpyrifos is commonly available and relatively inexpensive, it is likely to be the insecticide of choice when broadcast applications of contact insecticides are desired.

The down side of residual activity is that contact insecticides typically kill indiscriminately, so they will be taking out native ants as well as fire ants. We recovered few native ants on the highly effective bifenthrin plots. Under natural conditions, native ants can play an important role in keeping fire ants at bay.

Bait-station sampling seems to be a more accurate measure for assessing efficacy of fire ant insecticides. It is however, more difficult to perform and requires better trained field personnel. With further refinements to the process, bait-station sampling could be made easier and less expensive to perform and may become a new gauge for measuring fire ant insecticide efficacy.

Acknowledgments

Our thanks to: The City of Warren, Arkansas and the Monticello Airport Commission, who graciously provided land for the experiment; FMC Corp., who supplied the Talstar®; and American Cyanamid Co., for providing Amdro®. This research was funded by USDA, APHIS Grant 97-810-0229-GR, and this report does not necessarily express APHIS's views.

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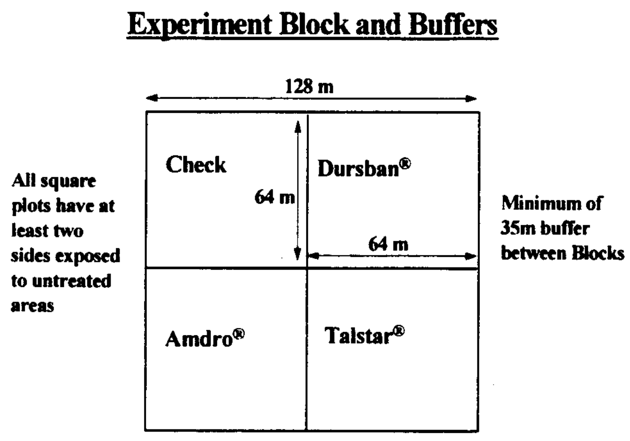

Fig 1. Layout of block with treatment plots.

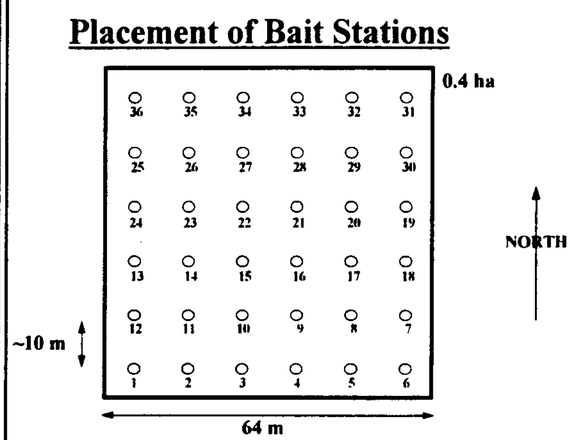

Fig 2. Bait station placement in each treatment plot.

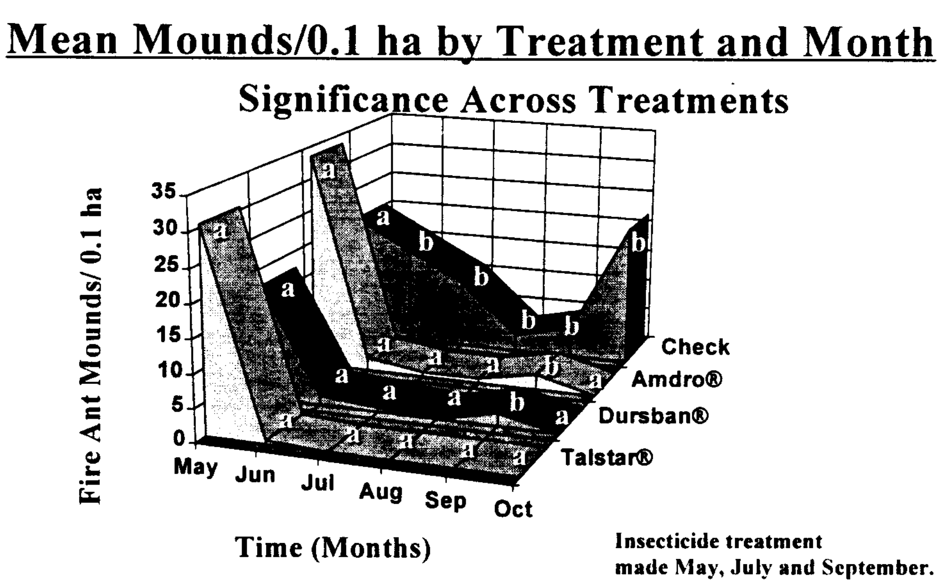

Fig 3. Imported fire ant mounds per 0.1 ha showing significance across treatments. Means with the same letter within a month are not significantly different at the 0.05 level, Tukey's Studentized Range Test

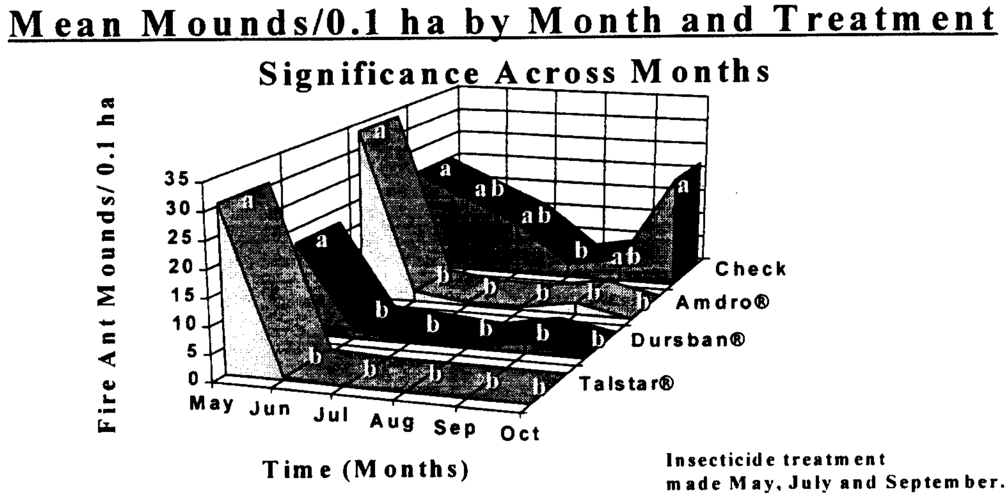

Fig 4. Imported fire ant mounds per 0.1 ha showing significance across months. Means with the same letter within a treatment are not significantly different at the 0.05 level, Tukey's Studentized Range Test

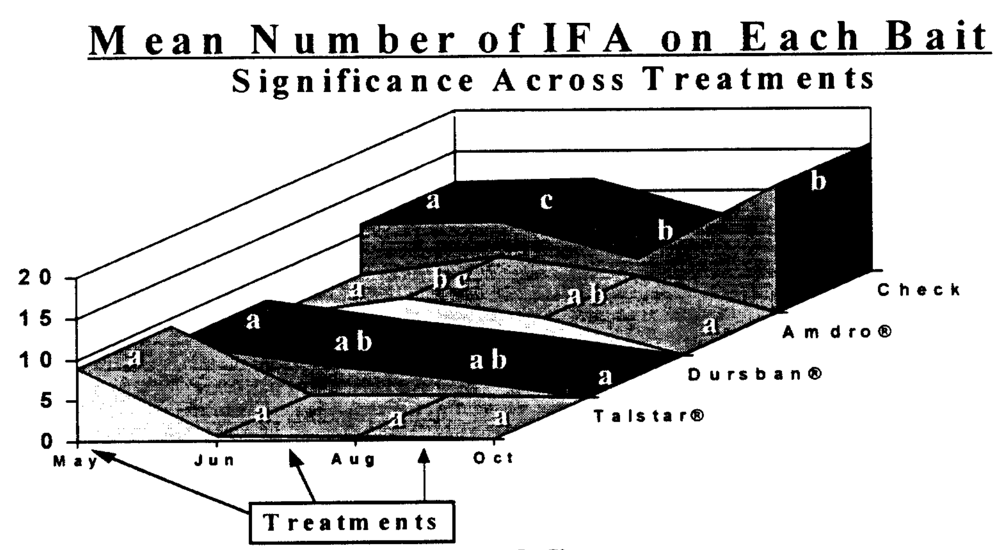

Fig 5. Mean number of fire ants present on each bait. Means with the same letter in the same month are not significantly different at the 0.05 level, Tukey's Studentized Range Test

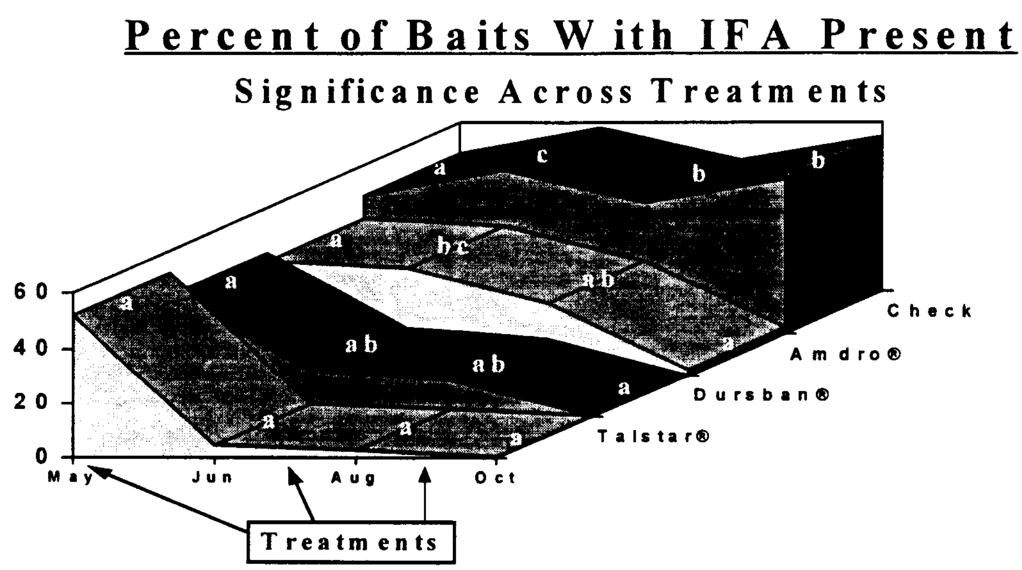

Fig. 6. Percentage of baits with fire ants present. Means with the same letter in the same month are not significantly different at the 0.05 level, Tukey's Studentized Range Test

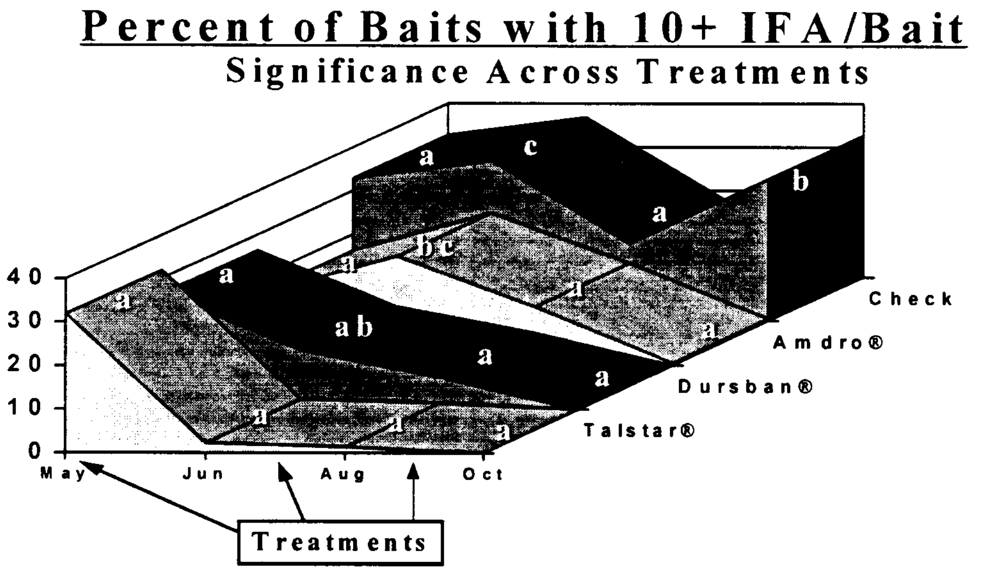

Fig 7. Percentage of baits with 10 or more fire ants present. Means with the same letter in the same month are not significantly different at the 0.05 level, Tukey's Studentized Range Test.

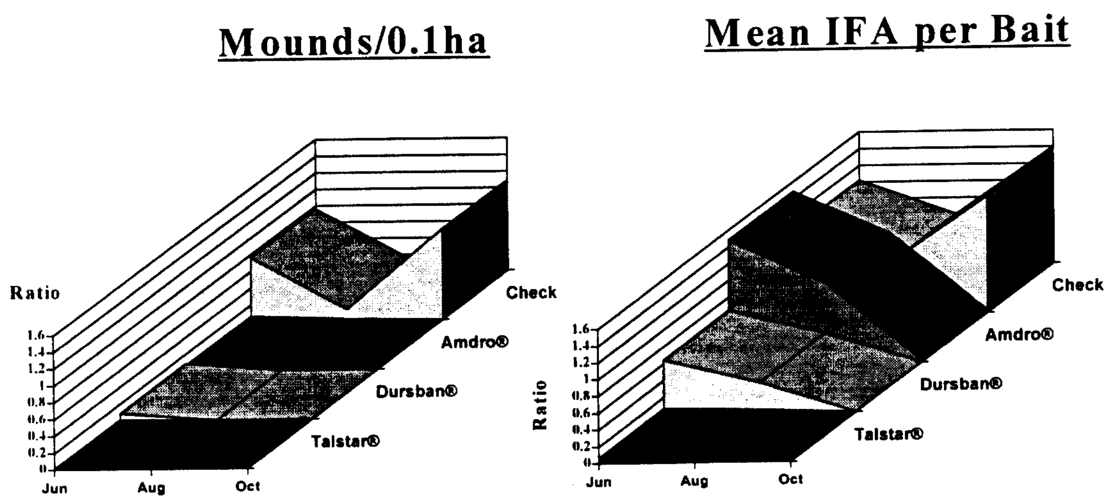

Fig 8. Comparison between mound counting and the mean number of fire ants collected at each bait expressed as a ratio to the pre-treatment levels (May) for each month and treatment.

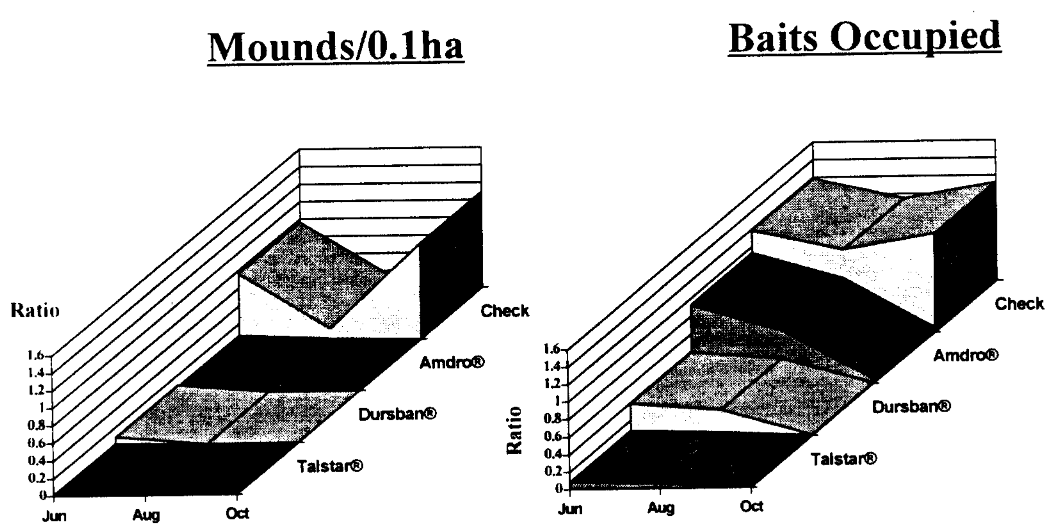

Fig 9. Comparison between mound counting and the number of baits occupied by fire ants expressed as a ratio to the pre-treatment levels (May) for each month and treatment.

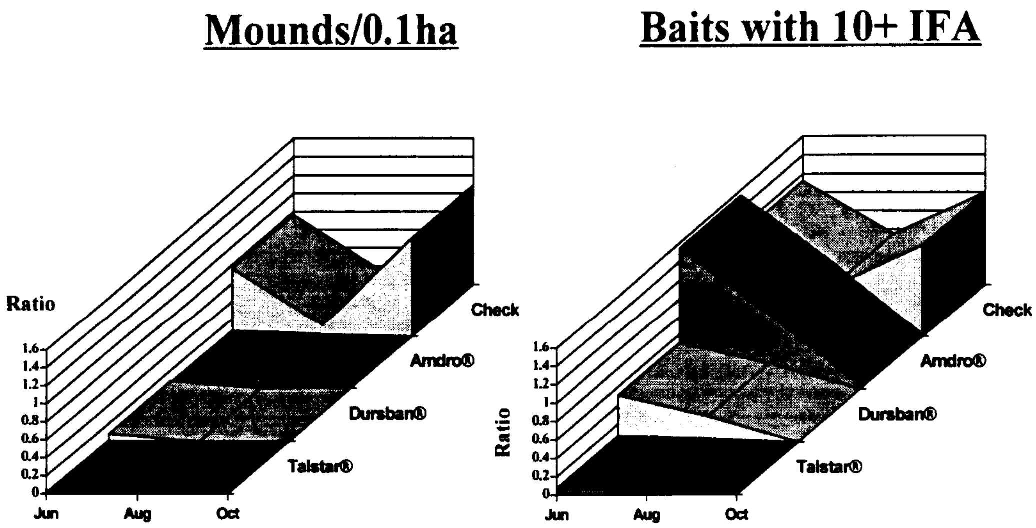

Fig 10. Comparison between mound counting and the number of baits occupied by 10 or more fire ants expressed as a ratio to the pre-treatment levels (May) for each month and treatment.

Impact of a Microsporidian Pathogen, Thelohania solenopsae, on Red Imported Fire Ants

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Thelohania solenopsae Knell, Allen, & Hazard is an entomopathogenic microsporidium that was first reported by Allen and Buren (1974) from alcohol preserved specimens of the red imported fire ant, ***Solenopsis invicta*** Buren, collected in Brazil. *T. **solenopsae*** infections have since been confirmed in the black imported fire ant, ***Solenopsis richteri*** Forel, in Argentina (Moser 1995). Spores of ***T. solenopsae*** are found in fat bodies of adults, and vegetative stages are found in the fat bodies of larvae and pupae, and in queen ovaries. Vegetative stages are also found in eggs, and thus *T. **solenopsae*** is transovarially transmitted (Briano ***et al.*** 1996). Briano and Williams (1997) reported negligible effects on the longevity of adult workers and sexuals of ***S. richteri.*** Effects of ***T. solenopsae*** on the development times of immature stages of fire ants are unknown. However, in a field study in Argentina, Briano ***et al.*** (1995) reported an 83% decrease in the density of black imported fire ant colonies within 4 years. Because of the apparent lack of an effect on adult workers, and the slow reduction in colony densities, it was suspected that ***T. solenopsae*** reduced brood production.

In 1996, Williams et al. (1998) discovered ***T. solenopsae*** infected ***S. invicta*** in several locations in Florida. Limited surveys throughout the southern U.S. revealed that infections were present in Texas and Mississippi. With the discovery of ***T. solenopsae*** in the ***U.S.,*** studies have been conducted on the impact of this pathogen on colonies of S. ***invicta.*** We report the effect of ***T. solenopsae*** on brood production in laboratory colonies, and the inoculation and infection of field colonies of S. ***invicta.***

Material and Methods

Laboratow Inoculation 1. Monogyne laboratory colonies, reared from newly mated queens, were used to examine the effects of T. ***solenopsae*** on brood production. These colonies contained an average of 71,000 workers (range: 50k-95k), 68 ml brood (range: 30-90 ml), and 1 queen. To inoculate a colony, 1 gram of brood (an arbitrary mixture of eggs, larvae, and pupae) was separated from field collected ***S. invicta*** colonies that were infected with T. ***solenopsae.*** The brood was placed next to a colony so that it would be carried into the colony by worker ants. A total of ten colonies were inoculated, and 1 gram of uninfected brood per colony was also introduced into ten control colonies.

The brood volume and the number of adult worker ants per colony were catergorized visually, using procedures that were adopted from Banks and Lofgren (1991). In addition, to assess the effect of ***T. solenopsae*** infection on queen weights, pre- and post-inoculation weights were recorded for each queen. Colony population estimates and queen weights were obtained before brood introductions, 8 weeks later, and at 2 or 3 week intervals thereafter for 23 weeks. Brood volumes, worker populations, and queen weights from inoculated and control colonies were summed over sample dates and compared by t-tests.

Laboratory Inoculation 2. To examine the effect of *T. solenopsae* on oviposition rates, a set of five laboratory fire ant colonies were inoculated with brood from *T. solenopsae* infected colonies and another set of five colonies had uninfected brood introduced. These colonies had an average of over 80,000 workers, 59 ml of brood, and 1 queen. Eight weeks after brood introductions, queens were removed from the colonies, and the number of eggs laid in 1 hour was recorded. Queens were then returned to the colonies, and the procedure repeated at 12, 16, and 21 weeks after brood introductions. Zero eggs were assigned to queens that died after 8 weeks. Queen weights were also recorded when brood was introduced and when oviposition rates were determined. Oviposition rates and queen weights from inoculated and control colonies were averaged per colony over sample dates ≥8 weeks after brood introductions and then compared by *t*-tests.

Field Inoculation. Brood from infected or non-infected *S. invicta* colonies were introduced into polygyne *S. invicta* colonies located in a pasture in Alachua County, Florida. Introductions were made by making an opening in a mound with a shovel and pouring 5 gm of brood into the opening. A total of five mounds were inoculated with infected brood and five control mounds had uninfected brood introduced. Adjacent inoculated mound were located within 29 feet of each other, and adjacent control mounds were within 45 feet of each other. Inoculated and control mounds were grouped in separate areas that were a minimum of 135 feet apart. Brood introductions were made on May 8, 1997.

Before brood introductions, colonies were checked for the presence of *T. solenopsae* by looking, under phase contrast microscopy, for spores in a supernate obtained from groups of about 50 worker ants that were macerated with water in a tissue grinder. On the 8th week after brood introductions, and at 3 to 6 week intervals thereafter for 40 weeks, adult worker and larval samples were obtained from each mound to determine if colonies were infected. Adult workers were examined for spores as described above. Larval samples (10 larvae per colony) were individually smeared onto a slide, stained with Giemsa, and examined for vegetative stages of *T. solenopsae*. On the 40th, 44th, and 48th weeks after brood introductions, adult worker samples from mounds surrounding the inoculated and control mounds were also examined for *T. solenopsae* spores to check for evidence of spread. To assess effects of *T. solenopsae* on colony populations, the USDA population index ratings (Lofgren & Williams 1983) were determined for inoculated and control colonies on each worker/larvae sampling date.

Results and Discussion

Laboratory Inoculation 1. Inoculated colonies had significantly lower brood volume (t = 6.67, df = 18, $P \leq 0.0001$), worker populations (t = 5.74, df = 18, $P \leq 0.0001$), and queen weights (t = 6.25, df = 18, $P \leq 0.0001$) than control colonies. Reductions in brood and workers were evident at 10 and 12 weeks after inoculation, respectively (Fig. 1 and 2). At the end of the study (week 23), there was an average of 6 ml brood per colony from inoculated colonies in contrast with 50 ml in the controls. Queen weight reductions in inoculated colonies were evident at the initial post-inoculation sample at 8 weeks (Fig. 3). The slow decline in brood and worker populations, along with the concomitant reduction in queen weights suggested that *T. solenopsae* caused a decrease in brood production in colonies.

<u>Laboratory Inoculation 2.</u> The average number of eggs laid per queen per hour from inoculated colonies was 66% lower than control colonies (t=2.03, df=7, P=0.0816) during the period from 8 to 21 weeks after brood introductions (Fig. 4). This period was compared because previous studies indicated that the effects of *T. solenopsae* infections generally became detectable 8 weeks after inoculation. Oviposition rates between inoculated and control colonies at week 0 were not significantly different (t= -1.151, df=7, P=0.2874).

Pre-inoculation queen weights did not differ significantly (t=-1.287, df=7, P=0.2389) between inoculated and control colonies. After brood introductions, average live queen weights were 18% lower in the inoculated colonies than in controls (t=1.457, df=7, P=0.1884). However, 3 out of the 5 queens from inoculated colonies were dead by week 16 and thus their weights were excluded from the average when they died. The lack of significantly lower queen weights from the inoculated colonies might be attributed to the death of queens in these colonies, and consequently, only heavier surviving queens were available for the queen weight average. All control queens were alive on the 21st week.

<u>Field Inoculation.</u> *T. solenopsae* infections were detected in four of the five inoculated colonies 18 weeks (Sept. 1997) after brood introductions. Both meiospores in adults and vegetative stages in larvae were recovered from these four colonies throughout the study. *T. solenopsae* was not found in any of the control colonies. After 11 months, all inoculated colonies were still active, however, there was a 30% reduction in the average population index for the inoculated colonies in contrast to a 4% increase in the controls.

At 40 and 44 weeks after inoculations (Jan. and Feb.1998), eight additional colonies surrounding the inoculated mounds were found to be infected with *T. solenopsae*. In week 48 (March 1998), *T. solenopsae* was found in 12 more colonies near the inoculated mounds. Infected colonies found in weeks 40 and 44 were still active and infected in week 48. However, because non-inoculated colonies that were later found to be infected were not confirmed to be uninfected by *T. solenopsae* prior to infection, evidence for the natural spread of *T. solenopsae* is not conclusive.

This is the first documentation of artificially initiated horizontal transmission of *T. solenopsae* infections among fire ant colonies. The laboratory inoculations showed significant reductions in brood and workers from infected colonies. Lower oviposition rates, queen weights, and queen survivorship was also documented from infected colonies. Reductions were first noticeable 8 to 12 weeks after inoculations in monogyne, laboratory colonies. Artificial inoculations resulting in infections of red imported fire ant colonies under field conditions was also demonstrated for the first time. After 48 weeks, a 30% reduction in colony populations was observed in the infected colonies. Since field inoculations were made in polygyne, *S. invicta* colonies, the impact of *T. solenopsae* may have been slowed or reduced because it is possible that all queens may not be infected. The ability to artificially infect *S. invicta* colonies with *T. solenopsae* should facilitate the assessment and development of this pathogen as a biological control agent of imported fire ants.

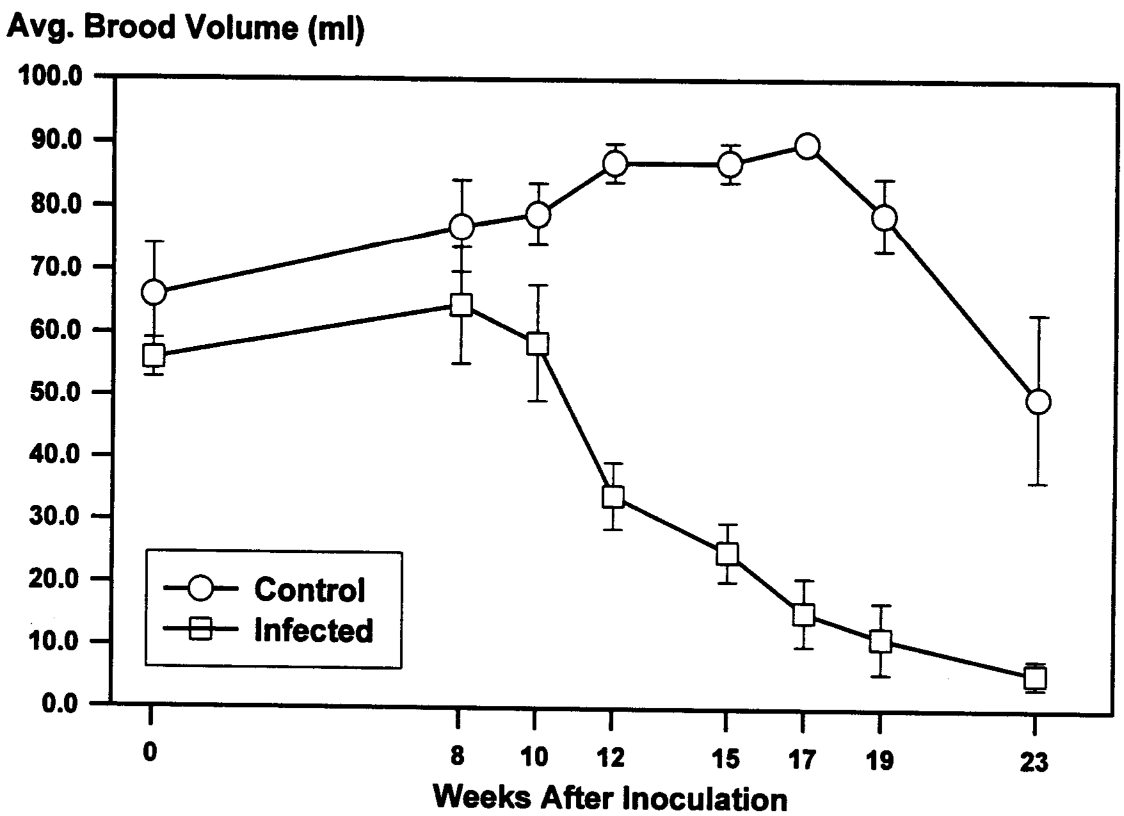

Fig. 1. Average (bar: ±1 SE, N=10) brood volumes from laboratory colonies of *S. invicta* provided with brood from uninfected (control) and *T. solenopsae* infected colonies.

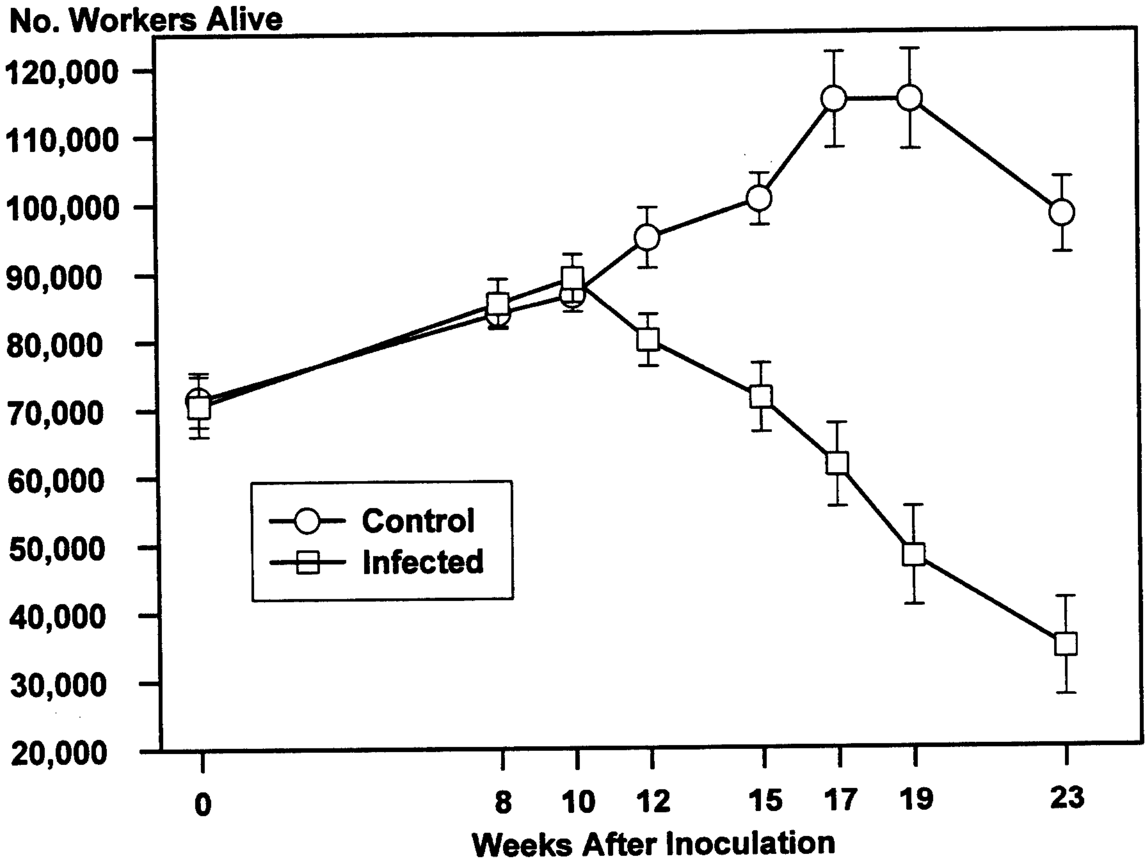

Fig. 2. Average (bar: ±1 SE, N=10) number of live adult workers from laboratory colonies of *S. invicta* provided with brood from uninfected (control) and *T. solenopsae* infected colonies.

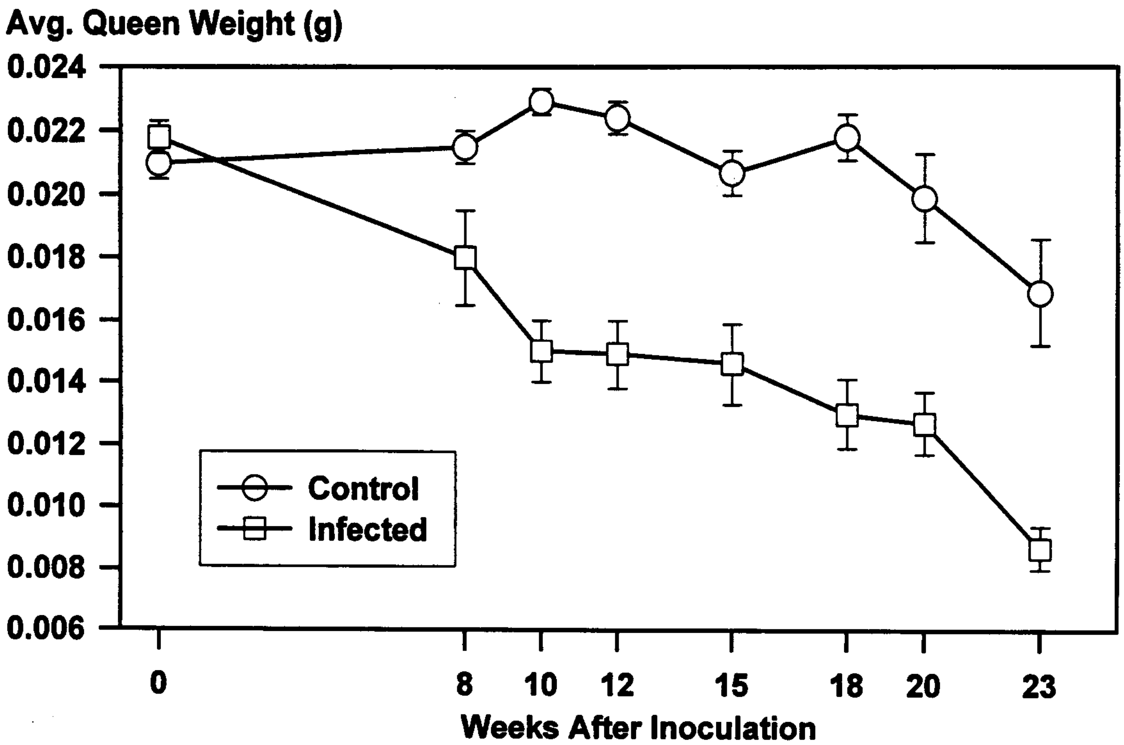

Fig. 3. Average (bar: ±1 SE, N=10) queen weights from laboratory colonies of *S. invicta* provided with brood from uninfected (control) and *T. solenopsae* infected colonies.

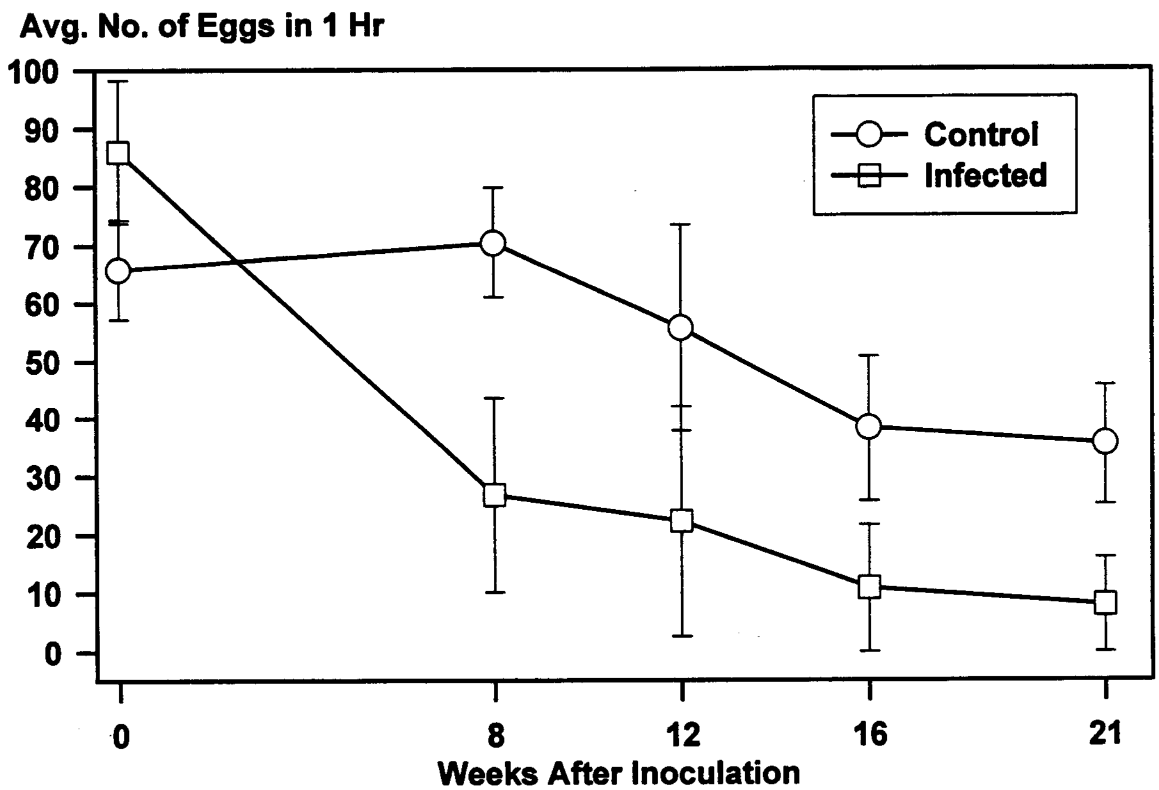

Fig. 4. Average (bar: ±1 SE, N=4 infected, N=5 control) oviposition rates from laboratory colonies of *S. invicta* provided with brood from uninfected (control) and *T. solenopsae* infected colonies.

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Analysis of Acoustic Communication by the Black Fire Ant

Solenopsis richteri

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Introduction

Communication is essential in highly organized ant societies and can occur via a diverse set of inter-related modes of communication. Much of the more recent work on communication has focused on chemical communication via pheromones (Wilson, 1971; Holldobler and Wilson, 1990) with less focus on visual communication or acoustic communication. While visual communication is not likely to be employed extensively by ants, acoustic communication may well prove to be as significant a communication mode as is chemical communication (Hickling & Brown, in review) and was considered an important communicatory mode by Wheeler (1910).

The first report of the stridulatory structure and stridulation in ants was by Landois in 1874 (Landois, 1874 in: Wheeler, 1910). Several other workers from the late 1800's investigated the stridulatory structure in a variety of ants, including representatives from Myrmicinae and the Ponerinae. The file was described as consisting of extremely fine transverse and parallel ridges at the base of the first gastric segment. While the ridges are generally of similar size, there is at least one known example from Australia (genus *Mima*) in which the file is divided into two parts. One of the parts has finely spaced ridges; the other has coarsely spaced ridges. Sharp (1893) noted that "a stridulatory performance by this insect might produce very extraordinary effects". In an extensive survey of 1,354 ant species in 205 genera, Markl (1973) demonstrated that a stridulatory structure is present in the subfamilies Ponerinae, Nothomyrmeciinae, Pseudomyrmecinae and Myrmicinae but is lacking in the most primitive subfamily. He found that, with few exceptions, ants that nest in the soil tend to have a stridulatory structure, and arboreal ants lack the structure.

While ants may stridulate in a variety of contexts (Stuart and Bell 1980; Spangler 1967; this study), the effects of the sounds on other ants have been investigated experimentally in a relatively small number of instances. Also, In relatively few cases have researchers demonstrated multiple contexts for sounds within a given species. Most stridulatory studies have been on adult ants, however, in a few instances, larvae are also known to stridulate or respond to stridulations (Wheeler and Bailey 1920; Spangler 1967).

Little is known about sound reception in ants. Ants are known to have well developed subgenual organs near the base of their tibiae (Wilson, 1971). These chordotonal organs, which also are present in other groups of insects, are known to be sensitive to vibrations through the substratum, but their sensitivity to air borne vibrations has not been demonstrated. Johnston's organ, also a chordotonal organ, perceives forces acting on the antenna of ants. Although this organ perceives sound in some insects, its function in ants has been speculated to be involved with orientation to gravity (Chapman, 1982). The presence and function of other chordotonal organs has not been demonstrated in ants, although such organs are common on the bodies of other insects.

There appear to be four primary functions of stridulation in ants: intraspecific alarm signals, recruitment signals, warning signals to potential predators, and signals during reproduction. In addition to these primary functions, stridulation has also been reported in *Leptogenys* during nest emigration (Maschwitz and Schonegge, 1977), trophallaxis and allogrooming in *Leptothorax muscorum* (Stuart and Bell 1980), and while drinking in *Pogonomyrmex* (Spangler 1967).

Alarm signals.- Several authors have noted than many species of ants will stridulate any time the ants are restrained. In the lab, it is possible to induce ants to stridulate by restraining them, and this can then be detected with a microphone. Stridulation plays a role in underground alarm communication when ants may be restrained by a cave-in of their nest (Markl, 1965; Markl, 1967; Spangler, 1967). In both the leaf cutting ants and in the harvester ants, workers will respond to buried nest mates and are stimulated to dig and extricate them. Spangler (1967) found that workers of *Pogonomyrmex* excavated a buried ant more quickly when stridulatory vibrations were present but did not orient to the sound. Apparently the sounds facilitated the digging behaviors.

Recruitment signals.- (Roces *et al.*, 1993) demonstrated that in the context of foraging, leaf cutting ants use stridulation to recruit workers to higher quality leaves. 100% of the workers stridulated while cutting leaves of high quality, but only 40 % stridulated while cutting leaves of lower quality. The context of the signal is important: the same species will respond with digging behavior when an individual is trapped underground and is stridulating.

The vibration of stridulation of the large foragers in the leaf-cutting ant *Atta cephalotes*, serves as a close-range recruitment signal to the minim workers (Roces and Holldobler, 1995).

The minims then "hitchhike" on the leaf fragment cut by the large workers and actively defend the leaf carriers against parasitic phorid flies. The same authors (Roces and Holldobler, 1996) found that stridulation also facilitated cutting the leaves via a vibratome-effect. They demonstrated however, that the mechanical facilitation of stridulation is an epiphenomena of the recruiting function in this instance.

Warning signals.-Ware (1994) showed that workers of a South African species of ant stridulated when they were restrained or when C02 or mammalian breath was directed at them. The ants did not respond with stridulation when pure nitrogen or air was directed at them. Ware suggested that the function of stridulation is a defensive or warning sound to potential vertebrate predators. This species is capable of stinging and has strong mandibles for biting. Through ablation experiments, Ware found that the C02 receptors were probably located on the antennae. Likewise, the African ant *Megaponera foetens* produced stridulatory sounds as a response to disturbance and to air and CO2 mixtures directed at them. In this species, there was no evidence of intra-specific communication (Holldobler *et al.*, 1994). Here also the authors suggest that stridulation is an aposematic warning signal to potential vertebrate predators.

In an experiment using wolf spiders and mice as predators, Masters (1979) demonstrated that stridulating mutild wasps were not held as long or as likely to be killed as were an aphonic group of the same species. Masters suggests that stridulation could be a generalized aposomatic acoustic signal indicating that the stridulating individual may be unpleasant to consume. Master's also suggests that stridulation may elicit a startle response in a potential predator. Since fire ants will stridulate anytime they are restrained, it is possible that it is an interspecific cue to a potential predator.

Signals during reproduction.- Investigation of stridulation in ants in the context of courtship and mating is of particular interest as many species of arthropods use sounds extensively in species and mate recognition. However, in ants, the study of courtship and mating sounds is made more difficult by the fact that many species mate in flight or at the tops of trees and can not be conveniently recorded. The discovery of the terrestrial mating leks in four *Pogonomyrmex* species in the southwest U.S.(Holldobler, 1976a; Holldobler, 1976b) provided a unique opportunity to investigate the use of sounds in mating (Markl *et al.*, 1977), and since these four species were sympatric, this was an opportunity to investigate the role of sounds in reproductive isolation. In these species, the sexuals emerge from the nest following a rain and fly to a lekking area (often a bush or a defined area of ground) where mating takes place. At the lek, females will typically mate with several different males and are sometimes besieged by a large number of males attempting to mate with them. Mating can only be successful when the female turns her abdomen in a particular way. Markl *et al.*, (1977) demonstrated that stridulation is not used by the male or female sexuals and is used only intermittently by the workers before the nuptial flights. At the lek, recordings were made of free ranging (and undisturbed) individuals and of individuals in containers. Mating activity is accompanied by continuous wing vibrations; however, in all four species no stridulation of the sexuals was reliably recorded before or during copulation. Stridulation was first clearly recorded when the female no longer was willing to mate. The authors called this a "female liberation signal." There was some evidence that once the female started stridulating, the males became less persistent in attempting to mate. Finally, stridulation also occurs when founding queens compete for burrows but it has not been demonstrated that stridulation affects the outcome of fights for burrows. There was no indication that sounds or vibrations were important in reproductive isolation of the four species studied.

The first report of the stridulatory structure and sound production in *Solenopsis richteri* was in 1995 at the Imported Fire Ant Conference, San Antonio, Tx. The purpose of this present study was to further document the sound production by *Solenopsis richteri* and to investigate the contexts in which stridulation occurred.

Methods

The stridulatory sounds of *Solenopsis richteri* were recorded both in the laboratory and in the field at the University of Mississippi. For the field recordings, a probe fitted with a Bruel and Kjaer 1/2 inch microphone was inserted into a mound. Recordings in the laboratory were obtained by using a small test chamber consisting of a double sound-proof box with windows. A plastic container with a portion of an ant mound was placed inside the double box contacting a custom made sensor that consists of a sensitive microphone mounted in a stethoscope head (Hickling *et al.*, 1997). Analysis of the sounds was via a speech analysis system by Kay Electronics.

Results

Stridulatory sounds were recorded from *Solenopsis richteri* in at least 4 different contexts: alarm, attack, distress and group activity (Fig. 1, 8 seconds of sound from each context is shown). In each of the contexts, the sounds were distinct and recognizable. Sounds from each of the behavioral contexts can be heard on the following web site: www.olemiss.edu/~hickling/.

The "alarm signal" (fig. 1a) was generated when a probe with a microphone was inserted into a fire ant mound. A large number of ants were stridulating simultaneously and the microphone was attacked by the ants. The stridulatory activity following such an "attack" lasted for about 10 minutes and then gradually died down. The activity sounds of a small group of ants recorded in the sound box in the laboratory is shown in figure 1b. Typically, as the ants move about a small chamber there would be seemingly spontaneous bursts of stridulation. The sounds produced when ants attacked a caterpillar is shown in figure 1c. The stridulation of one individual in particular was distinct and loud. It is possible that this may serve to recruit other workers to a food source. Finally, a "distress signal" is indicated in figure 1d. In this instance, a single ant was restrained by its antennae being caught at the edge of a dish. More details on the sound pressure curves and sound pressure levels is available in Hickling *& Brown* (In review).

Discussion and Conclusions

Clearly *Solenopsis richteri* has a stridulatory structure and stridulates in a variety of circumstances. The process of stridulation involved an up and down movement of the gaster. The scraper is on the post petiole and the file is on the posterior end of the gaster. In *S. richteri*, there are 40-50 ridges on the file. By using this one structure, the ants are able to make different sounds by changing the speed of the abdominal movement. These ants can also vary the duration of the signals and the sound pressure levels according to circumstance (Hickling and Brown, in review). Stridulatory sounds may therefore be a versatile signal used in a variety of circumstances, including intra-specific and inter-specific contexts. The sounds may act synergistically with other signals (Masters 1979) particularly chemical signals (Spangler 1967).

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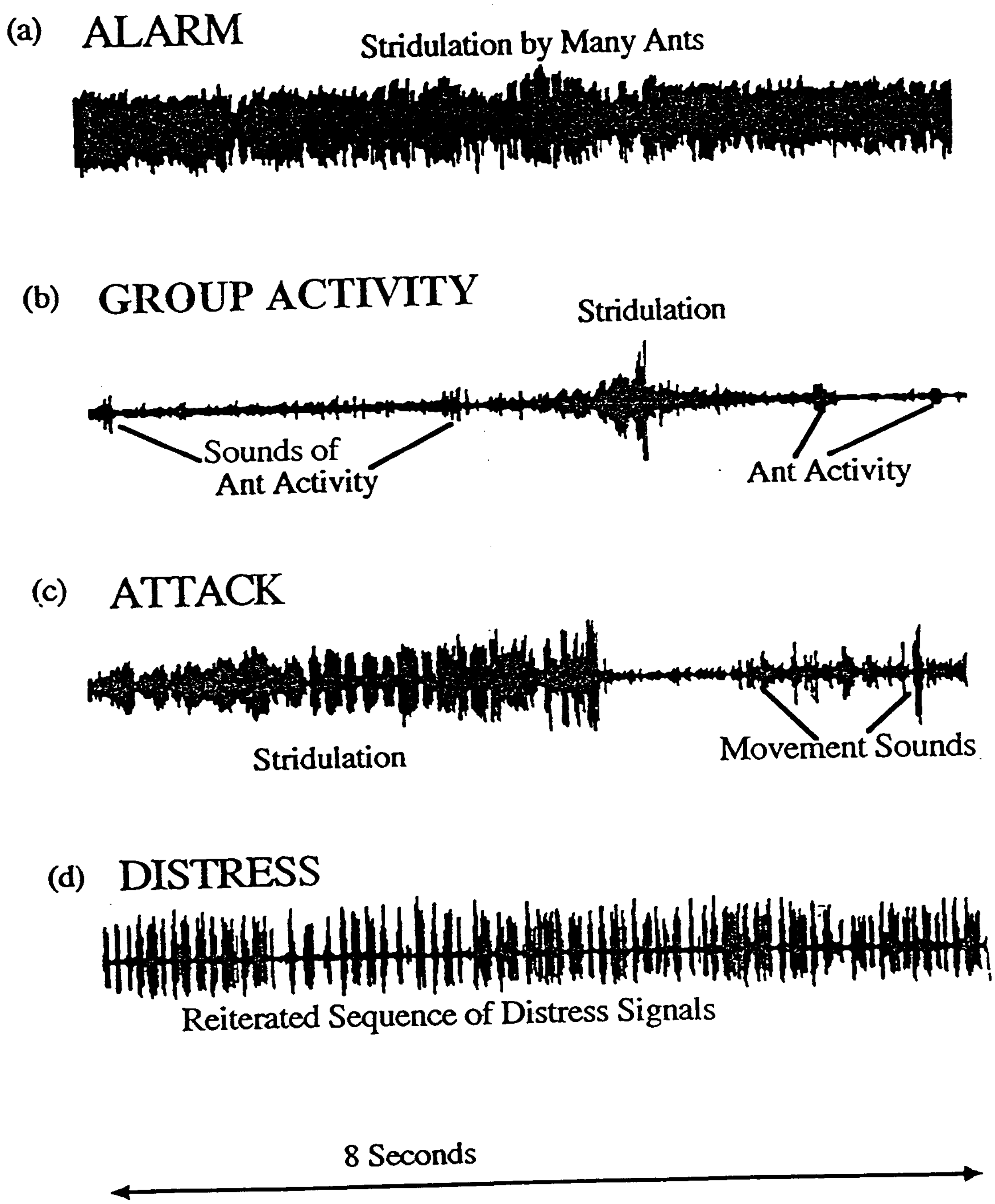
Fig. 1. Pressure-time traces of 8-second duration of acoustic signals emitted by *S. richteri* in situations of (a) alarm, when a microphone is thrust into an ant colony, (b) normal activity, (c) attack on a caterpillar and (d) distress signal from an ant with a caught antennae.



Desiccation resistance in the fire ant and its adaptability to arid environments

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Abstract

We used a quantitative genetic method to estimate genetic variation in desiccation resistance in a central Texas population of ***Solenopsis invicta*** **Buren**. We found significant genetic variation in time-to-death and the heritability estimate was significantly larger than zero ($h^2 = 0.42$). Head capsule width of the tested ants was measured as an indicator of their body size and its heritability was estimated as well. Body size and time-to-death was not genetically correlated, indicating that desiccation resistance is not a function of body size. Our results suggest that in a polygyne fire ant nest, queens with different degrees of resistance to desiccation may produce a cohort of workers that, as a group, can survive environments with a wide range of temporal and spatial variations in water availability.

Introduction

The red imported fire ant invaded the Unites States in the 1930's and became one of the most destructive pests of agriculture and urban environments in the infested regions (Callcott and Collins 1996). Habitat similarities between the newly-invaded, deforested southern US and the native South American flood plains (Vinson 1997) required little adaptation by S. ***invicta*** to its newly-invaded territory. The westward invasion of ***S. invicta*** into and across Texas was slowed in central and western Texas (Porter et al. 1991, MacKay and Fagerlund 1997) where the habitats may be significantly different from those previously occupied by the fire ant. Summer conditions in these Texas habitats may be drier and/or hotter than regions to the east.

Water availability in the environment is essential to survival and daily activity of arthropods (Block 1996). For fire ants to invade the drier environments bordering on the western edges of their home range, fire ant population must exhibit resistance to desiccation. Since a polygyne ***S. invicta*** colony consists of multiple queens that are not genetically related (Goodisman and Ross 1997), phenotypic and genetic variations of resistance to desiccation among fire ants in such a colony are likely to be greater than variations in a highly genetically related monogyne fire ant colony. Therefore, polygyne populations may be better able to respond to natural selection associated with dry areas of western Texas than monogyne populations. A recent study showed that fire ants from western Texas are more resistant to desiccation than those from eastern Texas (Phillips et al. 1996); however, the genetic basis for the difference was not examined.

The objectives of this study were (1) to examine the existence and magnitude of the genetic variation in S. ***invicta*** by estimating heritability of resistance to desiccation and of body size, and (2) to calculate the genetic correlation between the two aforementioned characters. Early research indicates that both monogyne and polygyne S. ***invicta*** queens effectively mate only once (Ross and Fletcher 1985). Therefore, we treated daughter ants produced by a wild-caught queen as full-sib sisters. Full-sib analysis was used to generate conservative estimates of heritability (Falconer 1981). We expected that genetic variation in desiccation resistance exists to some extent in the central Texas fire ant population.

Materials and Methods

Generating laboratory colonies of ants. Several hundred queens and thousands of workers were collected from 30 polygyne fire ant colonies along a roadside on the Texas A&M University Campus, College Station, TX. Two hundred and seventy artificial single-queen colonies were set up in the laboratory by placing one queen and 30 larger minor workers in a petri dish (100 x 15 mm diameter). The workers were marked with white spray paint to separate workers in the initial colonies from offspring generated by the queens. Petri dishes were ringed with Fluon (ICI Fluoropolymers, Bayonne, NJ) to prevent ants from escaping. A folded paper towel (3 x 3 cm, final size) was placed in a petri dish under the shade as an artificial nest. On alternative days the single-queen colonies were provided with water and fed a chicken egg-based artificial diet (Bhatkar and Whitcomb 1970), and occasionally the colonies were fed cut crickets and tobacco budworm larvae. Dead workers were replaced with new paint-marked workers as needed to maintain 30 workers per colony. Seven to 10 day-old daughters were harvested from the individual colonies and were subjected to desiccation tests.

Desiccation test and body size estimate. Desiccation tests were performed in a desiccation jar (9000 cm^3) filled with five pounds of $CaSO_4$ (W. H. Hammond DrieRite Co., Xenia, OH) dehydrated at 90°C for 12 hours. Temperature and relative humidity in the jar were measured continuously using a hygro-thermometer (VWR Scientific). During testing relative humidity usually dropped to about 19% while temperature remained around 25°C. Ants were placed singly in 0.2 mL PCR tubes that were taped upright onto the wall of the jar. Twenty holes smaller than the ant were made in the tube wall for ventilation. The ant activity was monitored hourly until they died. Time between test start and ant death was recorded. Body size of an ant was estimated using head capsule width measured on a dissecting microscope (4x power) equipped with an ocular micrometer.

Heritabilities and genetic correlation. Time-to-death data were normalized using square-root transformation before analysis. Variance components (V) of time-to-death and head width were estimated using the SAS VARCOMP procedure and covariance components (COV) were generated using the SAS NESTED procedure (SAS Institute, 1996). Heritabilities and genetic correlation were estimated using these variance and covariance components. Heritability (h^2) was estimated as

$$h^2 = \frac{\frac{4}{3}V_F}{V_P}$$

where V_F is the variance component due to families; V_P is the phenotypic variance consisting of V_F and V_E, which is the environmental variance. Because the fire ant is a haplodiploid species, V_F includes ¾ of the additive genetic variance (V_A), one-half of the dominance variance, a fraction of various epistatic variances, and total common environmental variance (maternal effects) (Li and Margolies 1993). Therefore, this heritability estimate sets the upper limit of the narrow sense heritability, V_A/V_P, which measures the degree of response to selection by a character (Falconer 1981).

A jackknife resampling method (Efron 1982) was used to estimate the standard error of heritability which is difficult to obtain using a standard method when data are unbalanced

(Falconer 1981, Li and Margolies 1993). The Jackknife estimates of heritabilities and standard errors are conservative compared to standard estimations (Li and Margolies 1993).

Genetic correlation, r_G, was estimated using the standard equations for single mating pairs which apply to both diploid and haplodiploid species (Li and Margolies 1993)

$$r_G = \frac{COV_{F(X,Y)}}{\sqrt{V_{F(X)}V_{F(Y)}}}$$

where $COV_{F(X,Y)}$ is the covariance component due to families, X represents one character and Y the other. The same Jackknife method used to estimate the standard error for heritability was used to estimate the standard error for genetic correlation.

Results

Fifty-nine percent of the single-queen colonies initially set up in the study produced one or more eggs within one week. Those that produced no eggs were discarded. In 62 families eggs developed successfully into adults, resulting in one to over 20 adult daughters per family during the test. Those that produced only one daughter were excluded from the sib-analysis, and no more than 20 daughters were tested from a single family.

Time-to-death and body size. All ants died within 34 hours under the test conditions. The average time-to-death was 16.3 hours and the mean head width of the ants was 0.57 mm (Table 1).

Table 1. Means, heritabilities (h^2) and genetic correlation (r_G) of resistance to desiccation and head width of *S. invicta*.

Character	Mean (±SD)	h^2 (±SE) (*P*)	r_G (±SE) (*P*)
Time-to-death	16.3 (5.5)	0.42 (0.13) (0.002)	-0.35 ± 0.20 (0.088)
Head width	0.57 (0.07)	0.54 (0.10) (0.001)	

Character means were based on 188 individuals from 41 families; Jackknife estimates of heritabilities and correlation were based on 41 families with an average of 4.5 daughter ants.

Heritabilities of desiccation resistance and body size and genetic correlation. We found a low but significant heritability of resistance to desiccation (0.42) and the heritability of body size (0.54) was also significant though relatively low compared to heritabilities of morphological characters in arthropods (see Roff and Mousseau 1987) (Table 1). There was no significant genetic correlation between time-to-death and body size (r = -0.35, Table 1).

Discussion

Desiccation is a strong environmental stress encountered by foraging fire ants in arid habitats. Fire ant workers in the College Station area stop foraging when temperatures reach

31 - 33°C on most surfaces but sustain their foraging at pond edges regardless of temperature (Martin, 1996). This finding indicates that water availability or micro-environmental humidity is vital to fire ant external activities. The significant genetic variation we found in desiccation resistance in *S. invicta* may have two significant implications. First, the fire ant is capable of responding to temporal or spatial fluctuation in water availability in the environment and adapting to drier habitats. The continuous acceptance of new queens year round by the polygyne fire ants (Goodisman and Ross 1997) will introduce new genetic variation in desiccation resistance during dry seasons. Second, a polygyne fire ant colony as a whole may remain active during extended day time periods because desiccation resistant workers can forage at times when it may be too dry and too hot for less resistant individuals. The variation, therefore, increases the survivability of the colony. This may represent a division of labor at a level only available or obvious in polygyne fire ant colonies. This division of labor furthers fire ant success in seasonally drier environments like central and western Texas.

Desiccation resistance is related to body size in non-social arthropods and ants when workers of all sizes are considered (Block 1996, Cerda and Retana 1997). Lack of genetic correlation between desiccation resistance and body size in this study indicates that variation of resistance to desiccation within a single worker caste largely reflects genetic differences among individuals and environmental influences. Our result is in line with the finding of Philips *et al.* (1996) that variation for desiccation resistance is not a function of body size of the workers tested.

The relative low heritability of desiccation resistance suggests that the ability of fire ants to further adapt to arid habitats is limited. Fire ant's westward spread will likely to be slow and could be slower, unless enough mutation in favor of surviving in this dry region accumulates. Given that the fire ant is a native species to humid regions or to wet habitats, adaptation to even drier conditions could take a long time. The low heritability may also indicate that there is not enough genetic variation in the polygyne fire ant to significantly enhance its survivability in dry or wet-dry fluctuating habitats. The advantage of polygyne over monogyne fire ants to survive in central and western Texas could be minimal, and the slow displacement of monogyne by polygyne fire ants observed by Greenberg et al. (1992) in College Station, TX area may reflect the slight advantage of polygyne over monogyne fire ants in this region.

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Comparison of Red Imported Fire Ant Populations In Texas Department of Transportation Rights-of-way and Adjacent Grasslands

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Vinson and Sorensen (1986) speculated that the clearing of highway rights-of-way (ROW) and then continued disturbance by mowing and maintenance would provide avenues for increased establishment of fire ants. Wilson (1971) noted that ***Solenopsis saevissima*** (Fabricius) built up very dense populations in open habitats but was largely absent from woodland. In northern Florida, **Tschinkel (1988)** reported ***Solenopsis invicta*** **Buren** more common along roadways than was ***Solenopsis geminata*** (F. Smith), and areas of high disturbance favored ***S. invicta***. Increased food, open canopy, and frequent disturbance made these habitats ideal for red imported fire ants.

Porter et al. (1992) surveyed roadsides in **Texas** and in Brazil for fire ants and reported that patterns of mounds in infested areas were generally unrelated to habitat and environmental conditions. For example, mound population densities were not well correlated with heights of grass along ROW (Porter et al. 1992). Direction of roads did not **affect** total numbers of mounds at sites nor numbers of mounds on each side of roads. Gross differences in solar exposure resulting **from** changes in road direction did not **affect** population densities of fire ant mounds along Texas roadsides (Porter et al. 1992). Roadsides in Brazil were generally not mowed; consequently, grass at sites in Brazil was about twice as high as in the U.S. However, no correlations were found between height of grass and mound population density among sites in either country. No significant relationships were found in several other environmental variables as well.

Additional work by Porter et al. (1997) compared the fire ant densities in different habitats between North America and South America. Porter et al. (1997) reported fire ant mound densities (**m^3** of mounds per ha) in North American mowed lawns to be 6.5 times greater than those in South America. Grazing areas contained 6.1 times more mounds in North America, and roadsides contained 5.3 times more mounds. Mound population densities in grazing areas and roadsides were not significantly different on either continent. Areas adjacent to crop land or residential areas did not contain unusually lower densities on either continent (Porter et al. 1997). When compared to the adjacent habitat type, no associations were found.

The Texas Department of Transportation **(TxDOT)** has been concerned that maintenance activities along highway ROW have encouraged the spread and establishment of red imported fire ants in Texas. The objective of this study was to determine if highway ROW harbored population densities of red imported fire ants that were **significantly different from** those of adjacent grasslands. **Significantly** greater population densities in ROW would indicate more suitable habitats for fire ant establishment and, consequently, a source from which adjacent properties could be infested. Potentially, ROW could act as dispersal comdors for continued spread of the red imported fire ant if they contained greater densities of fire ants.

Materials and Methods

Red imported fire ant populations were evaluated by an extensive sampling program along Texas highways and adjacent lands. Sites along transects were used to compare infestations across various habitat, geographic, and environmental gradients. Transects crossed through areas where red imported fire ants had been established for varying time periods in Texas. Transects were surveyed during May and June 1996. Transect sites were chosen by following state-maintained highways which transversed diverse vegetational areas of Texas (Hatch and Pluher 1993). Highways were of similar size, traffic intensity, and maintenance regimes. One north-south transect and two east-west transects through various Texas landscapes were sampled for fire ant populations and for ecological factors that may have impacted ants. A total of 28 sites were evaluated at approximately 80 km (50 miles) intervals. The north-south transect extended from north of Paris, Texas, on Highway 271 southward along Highways 19, 69, 59, and 288 to north of Lake Jackson, Texas. The southern east-west transect stretched from China, Texas, on Highway 90 westward along Highway 290 through Brenham, Giddings, Elgin, Austin, and Fredericksburg, and south to Junction, Texas, on Highway 377. The northern east-west transect extended from west of Texarkana on Highway 67, westward through Omaha, Mount Vernon, and Greenville, and along Highway 380 through Denton, Decator, and Runaway Bay.

Sites were selected on the basis of similarity in habitat between ROW and adjacent lands to provide uniformity for statistical analysis. Adjacent lands were of various usage types, but were consistently similar in that cultivated lands were avoided as were parking lots and recreational areas. Adjacent lands were typically grassland pastures. "Adjacent", therefore, refered to a tract of land immediately adjacent to ROW, "across the fence", and outside the maintenance regime of the Texas Department of Transportation. Sites were identified using both mileage details and global positioning (Geo Explorer, Trimble Navigation Limited, Sunnyvale, California).

At each site, numbers of red imported fire ants mounds were determined in three 0.10-ha plots in ROW and in the adjacent property. Mounds were rated for vitality following Harlan et al. (1981), and ratings were converted to the weighted system of Lofgren and Williams (1982). If a ROW site was too narrow to use a 17.85-m, rope-and-stake, circular plot method, the width of the site was measured, and 0.10 ha. was divided by the width to determine the length needed to equal the standardized area of 0.10 ha. Notations were made of the red imported fire ant mounds in ROW which were in contact with the roadbed ($\leq$1.0 m from the pavement).

Each site had two treatments. The highway ROW was one treatment, and the adjacent land was the second treatment. Mean number of red imported fire ant mounds in each treatment at each site was calculated and used in statistical analyses, and mean vitality ratings were calculated in each of the two treatment areas. Comparisons between the two areas were made using the Student's t-test (critical level; $P \leq 0.05$). Statistical analysis was done using StatWorks (Rafferty et al. 1985) computer software. Numbers for each transect were also pooled and evaluated by treatment (between location), and analysis of variance was used to test for differences among transects. All 28 sites were pooled together, and Student's t-test was used to test for statistical differences between locations. Typically, widths of ROW were 10.0 m; therefore, numbers of roadbed mounds to the remainder of ROW mounds were expected to be in the ratio of 1:9. This hypothesis was tested using chi-square analyses. Data were tested for normality using Kolmogorov-Smirnov one-sample test.

Discussion

Along Texas highway ROW, red imported fire ants were not more numerous than in the adjacent land areas. Mounds along roadbeds were smaller but more numerous than in the remainder of highway ROW. This finding does not agree with Porter et al. (1991) who reported no difference in numbers of mounds between ROW along edges of roadways and outer borders of ROW. However, Porter et al. (1991) surveyed areas 2.6 m from road surface, whereas the present study compared a 1.0 m width along the pavement's edge to the remainder of the ROW. From the present study, roadbeds may provide inoculum for invasion of the surrounding ROW and of adjacent land outside the areas of regular TxDOT maintenance.

Attraction of flying queens to reflective surfaces (Vinson 1997) may direct newly mated queens to paved road surfaces. Queens may crawl to adjacent roadbeds and find shelter beneath rocks and highway litter such as crushed beverage cans, paper, wood, cardboard, etc. The relative warmth provided by roadbed thermal mass (compare Thorvilson et al. 1992) and by moisture availability from condensation and/or runoff may furnish adequate habitat for founding queens. Small colonies that survive may find adequate food resources by scavenging animal materials on roadbeds (dead insects and larger animals) and by foraging in nearby mowed ROW.

Roadbeds may act as barriers for red imported fire ant colonies relocating within the ROW. A colony may butt against a roadbed, be prevented from moving across the pavement, and move along the roadbed. As colonies are flooded from ROW ditches during rain, they may drift against roadbeds and re-establish on drier ground. This re-establishment may account for high numbers of red imported fire ant mounds on roadbeds. As noted by Buren (1972) and Tschinkel (1986, 1988), red imported fire ants may be considered an opportunistic or early successional species dependent on ecological disturbance. In this study roadbeds may be a type of early successional habitat in that roadbeds contain more weedy plants and a different soil type than the surrounding ROW and adjacent areas.

Our study suggests that in areas of Texas where the red imported fire ant is well established, a population equilibrium of red imported fire ants between ROW and adjacent areas exists. Whereas other organisms have been observed to use habitat corridors within a matrix of unfavorable habitats (Haas 1995; Wegner and Merriam 1979; Dunning et al. 1995), red imported fire ants do not appear to be dispersing along the ROW in the areas studied. The highway roadbed does appear to provide a more favorable microhabitat as Thorvilson et al. (1992) found with brick walls and concrete structures in urban areas.

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each transect (Chi-square test, critical level $P \leq 0.05$, expected ratio 1:9, roadbed:remainder). More than the expected number of mounds were found in the roadbed within each transect. Mound numbers in roadbeds and in the remainder of ROW were not significantly different (ANOVA, LSD; $P \leq 0.05$) among transects. Analyses of pooled data from all 28 sites revealed significantly more mounds than expected in roadbeds (Chi-square test, critical level $P \leq 0.05$, expected ratio 1:9, roadbed:remainder).

Significantly more mounds were detected in the roadbeds than in the adjacent areas (Student's t-test; $P \leq 0.05$). In the adjacent areas the mean number of mounds per 0.10 ha was 23.0 (n=28; ±21.7 mounds); whereas in the roadbeds, the mean was 55.9 (n=28; ±64.9 mounds). Simple linear regression was used to determine if red imported fire ant mound population density in the roadbed was predictive of red imported fire ant mound density in the adjacent areas. The regression model was significant (F-value=4.7; P>F=0.04; slope=0.13; y-intercept=15.69); however, the coefficient of determination (r^2) was 0.15, indicating little of the variation in the adjacent area could be explained by the changes in mound density in the roadbed.

The mean vitality ratings of mounds within each transect were not significantly different between treatments (ROW vs. adjacent nor roadbed vs. remainder of ROW). When data from all 28 sites were pooled, mounds in the ROW reminder had significantly higher mean vitality ratings than did mounds in the roadbed (Student's t-test, $P \leq 0.05$), indicating that roadbed colonies were significantly smaller.

Bait cup collections within each transect did not differ significantly between locations (Student's t-test, $P \leq 0.05$). The southern east-to-west transect did have significantly fewer red imported fire ants per cup than did the other two transects (ANOVA, LSD, $P < 0.05$).

Scatter plots were constructed to compare vegetational analysis with red imported fire ant indices (mound densities and red imported fire ants per bait cup), and a linear regression model was fit to each scatter plot. Coefficients of determination measured the proportional change in red imported fire ant indices (mound density or red imported fire ants per cup) as determined by the variation in vegetation. This analysis was done for mounds in ROW and also in the adjacent areas, versus each layer of vegetation. Not all sites contained all layers of vegetation. Because grass and field were the dominant layers, they are reported herein. This regression analysis was also done for mean numbers of red imported fire ants per bait cup in each location (ROW and adjacent).

The best-fitting line for the regression model of numbers of red imported fire ant mounds per 0.1 ha and grass produced a coefficient of determination (r^2) value of 0.11 for ROW areas. In the adjacent areas the best fitting line (mound density versus grass) produced a coefficient of determination value of 0.06. Coefficients of determination for the layer of vegetation up to 2.0 m in height (grass + field layers) produced equally weak models. Significance levels for these data were greater than 0.05, except for mounds in the ROW and grass, which was 0.005.

Plotting the mean numbers of red imported fire ants per cup versus the various vegetational layers also failed to provide further understanding. Mean number of red imported fire ants per bait cup plotted against the grass layer produced a coefficient of determination of 0.02 in ROW and 0.01 in the adjacent areas. Plotting the mean number of red imported fire ants per bait cup in adjacent areas against the combination of the grass layer and the field layer (all vegetation up to 2.0 m) provided a coefficient of 0.04. Poor correlations between red imported fire ant indices and vegetation were found. Significance levels for these data were greater than 0.20.

Plastic cups (29.6 ml, Fill-Rite Corp., Newark, NJ), containing a protein bait (vienna sausage, Armour, Dial Corp., Phoenix, AZ), were placed at each site for approximately one hour. Cups were arrayed in three transects of 10 cups each, with 3.0 m between each cup and 10 m between each transect. These transects crossed through 0.10 ha circles that were surveyed for red imported fire ant mound population densities at each transect site. Both ROW and the adjacent land area were sampled at each site. Upon collection, bait cups were capped and stored on ice until specimens were identified and counted in the laboratory. Specimens were identified as either red imported fire ants or other ant species. Data represented recruitment of ants to the food source and provided an index of relative abundance of ants.

Mean numbers of red imported fire ants per cup, per site, and per transect were calculated. Mean numbers of other ant species were also calculated. For each site, the Student's t-test was used to compare mean numbers in ROW with those in adjacent land ($P \leq 0.05$). In the same manner as were the mound densities, the transect sites were pooled for comparisons, and all 28 sites were pooled for further evaluation.

Vegetation at each site was analyzed using a line-transect method for percent cover and was cataloged by physiognomic and structural descriptions (Kent and Coker 1992). Percent cover was recorded to the nearest five percent in each of the following classifications: bare ground, leaf litter, grass (vegetation 0.0-0.8 m in height), field (vegetation 0.8-2.0 m in height), scrub (vegetation 2.0-8.0 m in height), and canopy (vegetation over 8.0 m in height). Line transects ran through the 0.10 ha. circles. Six vegetation transects were surveyed at each location, each 20 m in length, and three transects were used in both the ROW and the adjacent land. Regression analyses was used to determine correlations between the numbers of red imported fire ant mounds and various vegetational characteristics measured at each site. CA-Cricket Graph III computer software was used in the regression analysis (Computer Associates 1992). Significance levels for the data were determined following Snedecor and Cochran (1980) by calculating the t-value from the correlation coefficient and the degrees of freedom in the model.

Results

Data along each transect varied with no obvious pattern emerging. Red imported fire ant mound population densities ranged from 0.0 mounds at some sites to 93.0 mounds per 0.1 ha. at one site in northeastern Texas. Recruitment of foragers to bait cups varied similarly. Vegetation was more consistent because of the research approach used to select study sites. Data were found to be normally distributed.

Pooled Transect Data. Data were pooled within transects, and the mean number of mounds within transects and among transects were compared. The greatest numerical mound density for ROW areas was that of the north-to-south transect which was 31.3 mounds per 0.1 ha. The highest mean mound density in adjacent areas was also along the north-to-south transect (28.8 mounds per 0.1 ha). Using Student's t-test, no significant differences were detected in the mean mound densities within transects between the ROW and adjacent areas. No significant differences among transects in mound densities in either ROW or adjacent areas were detected. When data from all 28 sites were pooled, analyses did not detect a significant difference among locations in mound densities (Student's t-test, $P \leq 0.05$).

Significant differences between roadbeds and remainder of ROW were detected within

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Scout dynamics and resource recovery by *Solenopsis invicta* Buren in experimental arenas.

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Abstract: Mature colonies of monogyne Solenopsis *invicta* Buren were placed in large arenas (6.5 x 1 m). The arenas were located in a glasshouse that exhibited diurnal temperature variation (cool mornings, hot afternoons, and warm evenings). The dynamics of foragers engaged in scouting behavior were observed with respect to the recent nutritional history of the colony, temperature, and humidity. The total number of ants engaged in scouting behavior was higher in colonies that were deprived of food for ca. 36-48 hours than when compared with colonies that were fed within the last 12-24 hours. In preliminary studies a significant regression between mean surface temperature and mean surface activity was observed under food deprived conditions ($n = 4$; P–0.012; r-square = 0.965) while no such relationship was observed under recently fed conditions ($n = 4$; P–0.6; r-square– -2.260).

Seed Predation by Fire Ants as an Alteration Mechanism of Plant Communities

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Abstract

Numerous studies indicate that many types of ants affect plant species composition and distribution in grasslands around the world. The red imported fire ant (Solenopsis invicta Buren) currently infests over 111 million hectares in the southeastern United States and is considered a major nuisance pest, yet there is little information on this species' effects on plant species composition or distribution. Results of a series of field and laboratory studies indicate that seeds of *Monarda* citriodora var. *citriodora*, a common aromatic annual forb, are collected by ants, not consumed, and discarded around the perimeter of mounds. The result is often a dense "ring" of Monarda plants growing around mounds the following growing season.

Field studies indicate that Ambrosia psilostachya and *Solidago altissima* may also form rings around fire ant mounds and be affected by ant foraging activities across larger areas. Other species, including Croton *monanthogynus*, Eustoma *granidiflorum*, *Bifora* americana, *Chaerophyllum* Tainturieri, Agalinis *purpurea*, and *Schizachyrium* scoparium var. scoparium showed differences related to mounds, disturbance type and/or the presence of foraging ants.

Introduction

Ants are known to have enormous influence on plant species composition in ecosystems around the world. Mechanism include seed distribution (Bossard 1991), seed protection from fire and rodents (Inouye 1980) (Harrington and Driver 1995) and differential soil and fertility characteristics of ant mounds (Green et al 1998). In a study potentially analogous to the red imported fire ant (Solenopsis invicta Buren) invasion of the United States, the Argentine ant (Iridomyrmex humilis) has displaced several harvester ant species critical to the maintenance of a fynbos shrubland in South Africa (Bond and Slingsby 1984).

Few studies have concentrated on what effects red imported fire ants may have on plant species composition in the southern U.S. This investigation first involved a large, long-term field trial in order to detect potential plant community changes in areas where fire ants were chemically suppressed versus left untreated and around their mound structures. Once differences were found, a series of laboratory and field trials were initiated to help elucidate the mechanisms involved.

Materials and methods

Field Study

The field study was conducted in two ungrazed pastures, referred to as the Lower Field and the Switchgrass Field, on land managed by the U.S. Army Corps of Engineers, Granger Lake, Williamson County, Texas. The soil in both fields was a heavy black clay classified as part of the Blackland Prairie of central Texas. The Lower Field was renovated from 1988 through 1991 in an effort to replicate a native tallgrass prairie. The Switchgrass Field was planted with native grasses around 1980. Switchgrass (*Panicum virgatum*) became the dominant species, reaching near-monoculture levels in spots. The Switchgrass Field was infested primarily by the monogyne form of RIFA, based on the observation of large mounds at densities between 50 and 751 hectare. The Lower Field contained primarily polygyne colonies, based on small mounds at densities of 135 to 3251 hectare (Porter et al. 1991).

The experimental design was randomized block with 4 replications with nested sub-plots. In the summer of 1993, the Lower and Switchgrass Fields were divided into eight 0.47 ha and 1.0 ha square Treatment Areas (TA's), respectively, so that each TA would contain at least 10 active RIFA mounds. Half the TA's were treated with a fire ant control bait product (either Logic®, Amdro® or a combination) at approximately 6 month intervals for the duration of the test to eliminate ant activity in marked mounds and suppress foraging within the TA's.

Within each TA, active RIFA mounds were permanently marked with steel reinforcing rod. Untreated TA's contained 10 mounds, to compensate for natural mound abandonment, while TA's to be treated with bait contained 6 mounds. Each mound represented the center sub-plot (abbreviated MD) of a cluster of 3 sub-plots. The remaining two were established at random points along a radius of 1.5 m from the center of each mound. One was left as an undisturbed control sub-plot (abbreviated CK). The third one was made to resemble an artificial mound or disturbed site (abbreviated DIST) by removing the vegetation, adding soil from a nearby spot and molding it into the shape of a fire ant mound. Each sub-plot in the radius was 28 cm in diameter and was marked with two nails as was the central mound so that they could be relocated with a metal detector over time.

Evaluation of plant species composition was conducted in June and October-November, 1994 - 1995. The test was discontinued in 1996 due to a severe drought. Evaluations were conducted by placing a belt quadrat sampler in the center of each subplot then identifying and counting all live plants rooted within each quadrat. The sampler consisted of two parallel steel rods, spaced 5 cm apart, embedded in a block of hardwood. The rods were marked in 10 cm increments to a length of 60 cm. so that each belt consisted of six, 5 x 10 cm quadrats.

In addition to this vegetation evaluation method, beebalm, *Monarda citriodora* var. *citriodora,* was evaluated by counting the number of plants rooted within 10 cm-wide concentric rings on and around RIFA-mound subplots out to 60 cm.

Laboratory seed preference and predation studies

Eight active RIFA colonies were collected 3 February 1997 from a cattle pasture near the Brazos River in Brazos County, Texas. The colonies were separated from the soil and kept in plastic sweater boxes coated with Fluon®, a Teflon-based emulsion that prevents insect escape. The colonies were fed only sunflower seeds and dilute honey-water in an attempt to acclimatize them to a diet of vegetable matter.

On 10 February 1997 seven colonies were selected for predation studies due to their aggressive feeding habits. Ten seeds each of beebalm (*Monarda citriodorai var. citriodora*), sunflower *(Helianthus spp.,* hulled commercial birdseed, used as positive control), and thistle (commercial birdseed, used as negative control) were placed in petri dishes. Three gray and three brown seeds of Croton (*Croton monanthogynus*) were also placed in the dishes. Seed were placed within six wedge-shaped dividing lines drawn on the bottom of the dishes so that seed movement, as well as removal, could be monitored.

The petri dishes were placed in empty colony boxes, referred to as foraging boxes, adjacent to the colony-containing boxes. The two boxes were connected at the beginning of the test by means of a bridge made from galvanized sheet metal. Each bridge was approximately 5 cm wide and 40 cm long. The metal was covered with packing tape to avoid ant contact with the potentially toxic galvanizing and to provide better traction. The number of seeds moved and removed were counted at 0:15, 0:30, 1:00, 2:00, 4:00 and 24:00 hours post-exposure.

The trial was repeated on 12 February. Seeds were kept on moist filter paper for 24 hours prior to ant exposure and kept moistened for the first four hours of the trial. Due to removal of some thistle seeds in the first trial, fungicide-treated cauliflower seeds were substituted as the negative control. After each test, seeds were recovered from all boxes and examined under a dissecting microscope for signs of damage.

A third trial was begun on 18 February 1997 to test whether RIFA would carry seeds farther than a few centimeters. Three particularly aggressive RIFA colonies were selected for this test. Each pair of colony and foraging boxes were separated by the length of two laboratory tables (each 0.75 x 1.1 m) and a metal bridge placed in each. To connect the boxes, a wooden board, 1.9 x 3.8 cm in cross-section and 2.44 m long was rested on top of the bridges so that it would not contact the box edges and allow ant escape. Empty colony boxes were used to fill the gap between the colony and forage boxes to further limit ant escape and catch any dropped seeds.

Ten each of sunflower and treated cauliflower (*Brassica* spp.) seeds and 100 beebalm seeds were placed in petri dishes in foraging boxes as before. The number of seeds removed was recorded at one, two, four, and 24 hours. The test was repeated on 20 February using pre-moistened seeds. Two weeks later, the colony and forage boxes were carefully examined and any beebalm seeds removed. These seeds were examined microscopically for ant damage.

Field seed predation

Based on results from the Field Test and Laboratory Seed Predation Tests, seeds from six species were selected for use in the Field Seed Predation Test: *Monarda citriodora*, *Bifora americana*, *Chaerophyllum Tainturieri*, *Palatina texana*, sunflower and cauliflower. These species were chosen because *Monarda, Bifora, Chaerophyllum,* and *Polytaenia* were in great abundance in the Lower Field at the time the test was performed, with all indicating possible positive correlations with ant mounds in results from the Field Test. Sunflower was chosen as a positive control to confirm foraging ant presence since it is highly preferred by RIFA and was easily available commercially. The cauliflower seeds were included as a negative control since RIFA sometimes manipulate and move seeds and debris for unknown reasons. As in the laboratory tests, it was hoped that the fungicide-coated seeds would discourage random investigation by the ants and give a better measure of the actual attractiveness of other species. Seed trays containing six shallow, round depressions arranged in a circle were used to offer the seeds. Each tray received seeds of all six species and each depression received 10 seeds of a single species.

In the Lower Field, approximately 1.7 kg/ha of Amdro® was applied to 4 treated TA's on 28 March 1997 by means of a gasoline-powered Solo® backpack blower modified to apply RIFA baits. Untreated TA's from the Field Test were used as untreated controls in this test, as well. On 14 July 1997, an active mound was located in each of the four untreated TA's and marked with a surveyor's flag. An additional random spot was marked in approximately the middle of each treated TA. To provide a level spot on which to place the seed-containing trays, the vegetation at each radius was trimmed down to bare ground using a gasoline trimmer equipped with a metal brush blade. Each bare spot was approximately 20 cm in diameter.

Beginning at approximately 7:45 a.m., 15 July 1997, the pre-filled seed trays were carefully carried into the field in a plastic holding box and one tray placed in each bare spot. Trays were secured by pushing a 16d nail through the pre-punched center of the tray into the ground. One randomly selected tray at each distance was covered with a 15 cm square x 4.5 cm tall cage made of 0.64 cm galvanized wire mesh to exclude rodents and birds. Cages were weighted with small rocks found nearby.

The first seed count was begun at 9:00 a.m. (one-hour exposure) and concluded at approximately 9:30. Succeeding counts were begun at 10:00 a.m. (two-hour exposure) and 12:00 noon (four-hour exposure). The number of seeds removed from each depression was recorded on pre-prepared data sheets. The final count was begun at approximately 7:30 a.m., 16 July (24-hour exposure). The nails and/or wire cages were carefully removed from each tray and the trays placed in a plastic holding box.

Results

Field Test

Numerous statistically significant correlations were found between several plant species and either RIFA mounds or the presence of ants. These findings are summarized in **Table 1**. Most of the findings were either too inconsistent or too weak to be considered definitive. However, the relationship of beebalm to fire ant mounds, particularly those in untreated areas, was consistent, very strong, and easily visible in the field.

Beebalm, an herbaceous annual that begins growth in March, then flowers and dies by July, is common in open pastures across Texas. The variety found in the study area is also known as the source of "citronella" insect repellent oil. Beebalm forms wide rings *around* RIFA mounds, extending from the edge out 20 - 30 cm. **Figure 1** illustrates the distribution of beebalm around all 3 sub-plot types in the June 1994 and 1995 evaluations in the Lower Field. Results were similar in the Switchgrass Field. Numbers are significantly different ($P<0.05$) out to 30 cm. **Figures 2 and 3** illustrate the mean number of beebalm plants found in 10 cm-wide concentric rings around marked RIFA mound sub-plots in the Lower and Switchgrass Fields, respectively. Note how the peak of each curve occurs about 15 cm away from the edge of the mean mound radius. The Switchgrass Field had larger mounds than the Lower Field, due to its monogyne infestation. **Figure 4** shows an "index" of beebalm population within 10 cm wide rings around marked mounds. Also of note was the complete absence of "rings" of beebalm around some mounds, while others had high concentrations. With all mound sub-plots taken together, 57% of them had beebalm rings, defined as 5 or more plants within 60 cm of mound center. The rest had virtually none. These results suggested that the mechanism by which the rings occurred was seed collection and incomplete consumption. Beebalm was, therefore, chosen for the all the seed predation studies that followed.

Laboratory seed preference and predation

The first set of seed predation studies indicated that sunflower was highly preferred by RIFA, while beebalm was only secondarily preferred. **Table 2** shows the results of the seed predation trials. Though sunflower seeds appear to have been completely consumed and any other seed largely ignored, beebalm was carried back to the RIFA colony boxes over a rather long distance. **Table 3** shows the fate of the beebalm seeds that were recovered after having been carried 2.4 m by RIFA.

Microscopic examination of recovered seeds indicated a few abrasions on one end of the seed coat, but no other damage. Also, no seed halves were located suggesting that the ants were unsuccessful in consuming any seeds. Germination rates were similar among recovered seeds than from a general sampling of the seed batch used in the tests (77%).

Field seed predation

Results of the field seed predation trial were consistent with those obtained in the laboratory. Sunflower seeds were highly preferred with all but 11 seeds of the 480 offered being removed within one hour. Beebalm was removed at a slower rate, but the numbers removed were significantly greater than those of non-attractive species. **Table 4** summarizes the results of the field seed predation study.

Discussion

Though populations of several plant species were suspected of being influenced by fire ants, it was only *Monarda citriodora* var. *citriodora* for which a likely mechanism could be proposed. It appeared that beebalm seeds are attractive in some way to fire ants who collect them and return them to the mound. The seed coats are too tough to be opened by the ants, who then discard or cache the seeds in close proximity to the mound. The following season, these seeds germinate around the usually-abandoned mound structure.

This idea fits with the visible pattern of beebalm growth - some mound structures had "rings" of beebalm and some didn't while virtually every "ring" of beebalm had a mound structure associated with it. It seems very likely that the following pattern occurs. Ants collect beebalm seeds during the early summer and discards them near a mound. Over the intervening months, the colony abandons the mound and re-locates. The next spring, beebalm seeds germinate, creating a "ring", while the active mound has no or few beebalm plants associated with it. In other words, for a mound to have a "ring" it must have been active during the seeding period of beebalm the previous year. A less-apparent "ring" may appear the following year on the original mound site due to ungerminated seeds, while a full ring would appear around the mound site that was occupied that second summer.

The question then remains of *why* ants would collect apparently useless seeds. Though by no means an answer, an intriguing hypothesis is suggested by Bishop and Thornton (1997). Their research shows that essential oils of *Monarda citriodora* var. *citriodora* have substantial anti-fungal properties through both contact and vapor action. Fire ants are known to have a host of fungal pathogens (). It is possible that there exists a symbiotic relationship between fire ants and beebalm. Fire ants benefit by the suppression of fungal growth by root exudates of beebalm and beebalm benefits through seed distribution to protected or more suitable germination sites. Given the short time that *Solenopsis invicta* has been in the study area, it is unlikely that such a relationship evolved between these two particular species. However, the attractive component of beebalm seeds could have evolved to attract the tropical fire ant (*Solenopsis geminata*) or other ant species displaced by the fire ant invasion.

Though the phenomenon of beebalm associated with fire ant mounds was the only definitive relationship established by these studies, there are likely many more species-specific fire ant-plant interactions occurring throughout the ants' range.

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Table 1. Summary of species abundance, distribution and possible mechanisms.

Species	Where most abundant	Possible mechanism(s)
Monarda citriodora	broad rings around mounds in untreated areas	RIFA seed collection
Ambrosia psilostachya	disturbed sites and mounds in untreated areas	disturbance, mound fertility/salinity
Bifora americana - Chaerophyllum Tainturieri	on and around mounds in treated areas	disturbance, mound structure
Agalinis purpurea	undisturbed sites in treated areas	disturbance
Eustoma grandiflorum	disturbed sites in untreated areas	disturbance
Solidago altissima	around mounds in untreated area	secondary predation / protection of phytophagous insects
Schizachyrium scoparium	mounds and in treated areas	disruption of nutrient cycle

Table 2. Mean number of seeds moved/removed from petri dishes in laboratory tests.

Hours post-exposure

Dry seeds - 0.3 m distance - 7 colonies

Seed	Init.	0:15	0:30	1:00	2:00	4:00	24:00
Sunflower	10	8.4/7.0	9.4/8.6	10/9.4	10/9.4	10/9.6	10/10
Beebalm	10	0.1/0	0.9/0.4	3.3/1.7	4.0/2.4	5.9/4.3	9.0/8.7
Thistle 10	0/0	0.4/0	1.7/0.6	3.0/1.7	3.6/2.0	3.4/2.4	

Moist seeds - 0.3 m distance - 7 colonies

Seed	Init.	0:15	0:30	1:00	2:00	4:00	24:00
Sunflower	10	3.7/1.3	6.3/4.7	7.6/6.0	9.3/9.1	10/10	10/10
Beebalm	10	0/0	3.7/1.7	6.4/4.3	9.4/8.3	10/9.6	10/10
Cauliflower	10	0/0	0/0 0.7/0	1.0/0	1.0/0.1	9.9/2.6	

Dry seeds - 2.44 m distance - 3 colonies

Seed	Init.	1:00	2:00	4:00	24:00
Sunflower	10	8.3/7.7	10/10	10/10	10/10
Beebalm	100	16.3/0	40.7/26.0	42.0/26.0	85.7/65.7
Cauliflower	10	2.0/0	3.7/0	4.0/0.3	5.7/0.7

Moist seeds - 2.44 m distance - 3 colonies

Seed	Init.	1:00	2:00	4:00	24:00
Sunflower	10	1.0/0.7	6.3/3.3	10/10	10/10
Beebalm	100	0/0	23.3/0	75.3/14.7	95.3/64.3
Cauliflower	10	0/0	0/0	0/0	3.3/1.3

Table 3. Fate of seeds removed from dishes in long distance predation tests

Colony	Forage box	Colony box	Total	Colony box seeds germinated
	Number of Beebalm seeds			
Colony 1	39	1	40	(seeds spilled)
Colony 2	44	24	68	14 (58.3%)
Colony 3	33	38	71	26 (68.4%)

Table 4. Mean number of seeds removed per tray depression by species

Species	1:00	2:00	4:00	24:00
	Hours post-exposure			
Sunflower	9.8[a]	10.0[a]	10.0[a]	10.0[a]
Cauliflower	0.6[c]	0.9[c]	1.2[c]	3.8[c]
Bifora	0.0[c]	0.02[c]	0.08[d]	1.1[d]
Chervil	0.0[c]	0.0[c]	0.04[d]	0.5[d]
Texas Parsley	0.0[c]	0.0[c]	0.0[d]	0.0[d]
Beebalm	1.9[b]	3.1[b]	3.8[b]	6.5[b]
F:	371.14	253.49	222.68	138.21
Prob:	0.0001	0.0001	0.0001	0.0001
R^2:	0.8681	0.8180	0.7979	0.7102
N=48				

Means followed by different letters in the same column were significantly different ($P<0.05$) using PC SAS PROC ANOVA and Tukey's studentized range test for mean separations

Figure 1. Beebalm distribution around plot types, 1994-1995, in the Lower Field

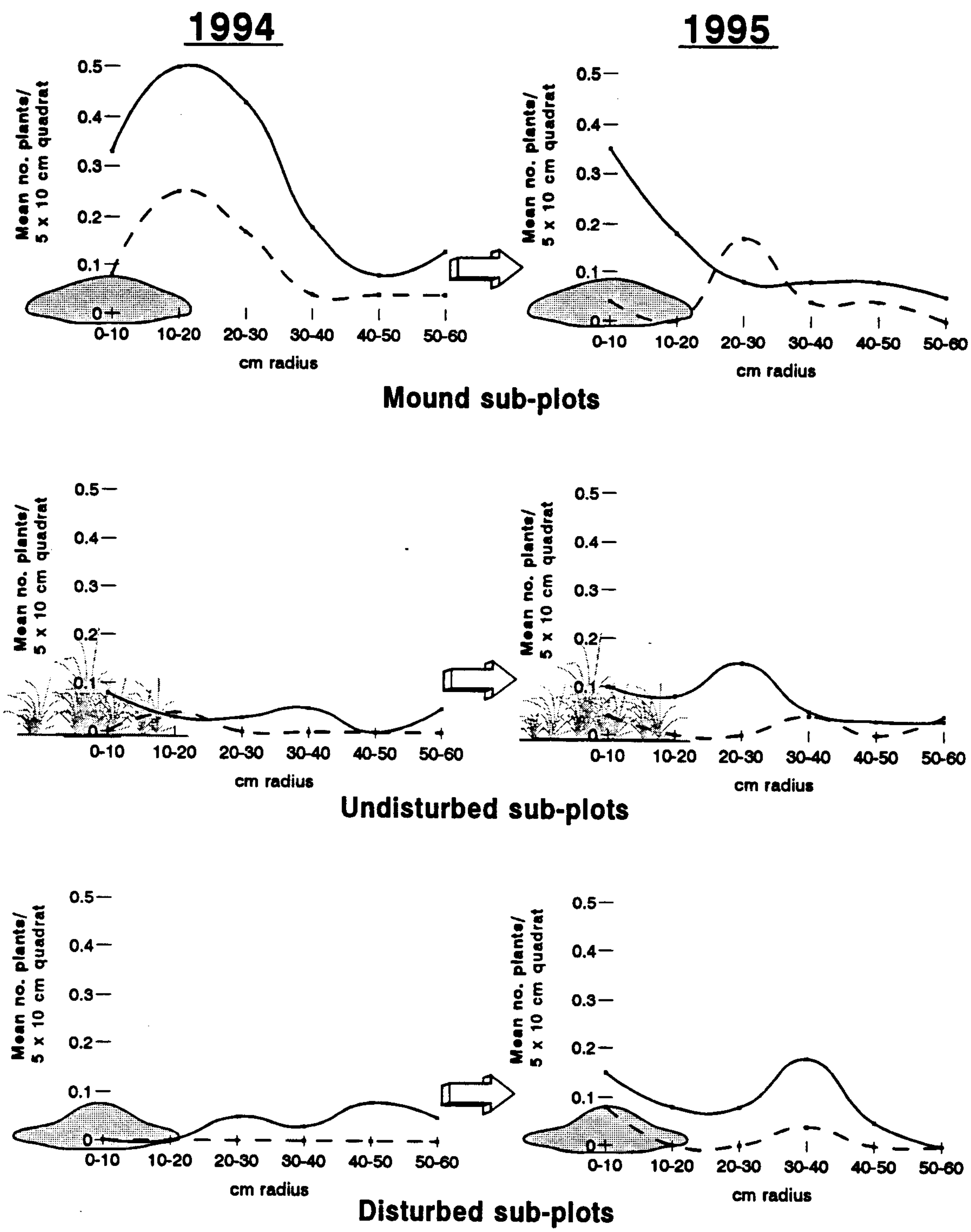


Figure 2. Beebalm distribution around mounds in Lower Field (polygyne).

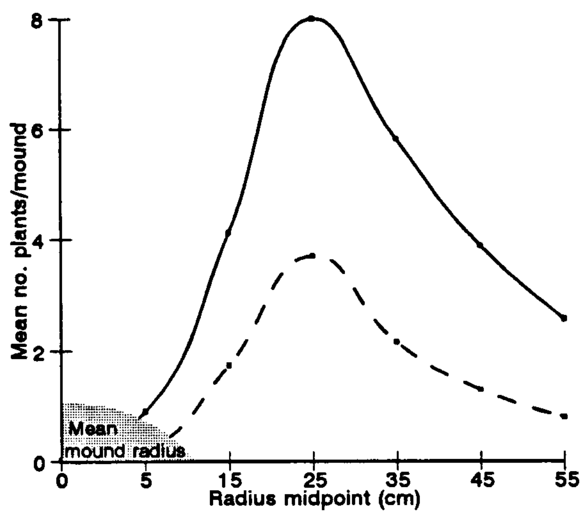


Figure 3. Beebalm distribution around mounds in Switchgrass Field (monogyne).

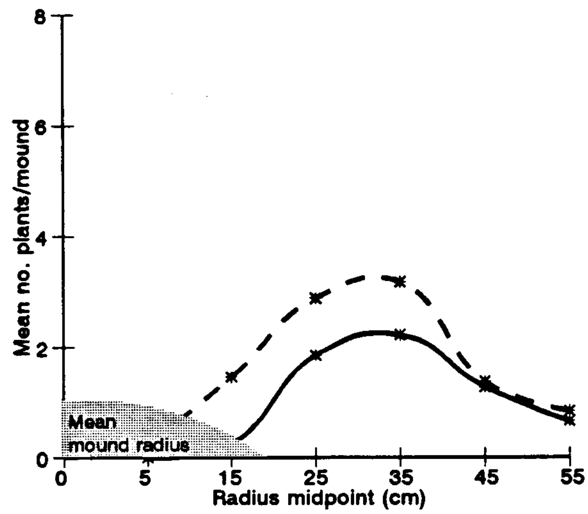
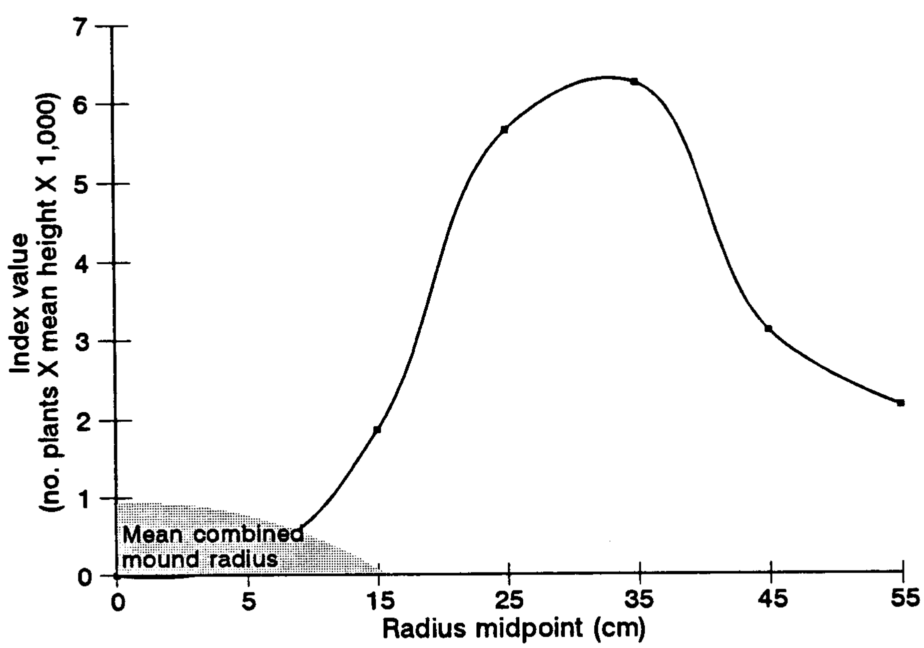


Figure 4. Beebalm population "index" of all mounds in both fields.



Newly Mated Queen Adoption & Polygyny

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Abstract:

There are several ways in which polygyne Solenopsis *invicta* colonies could originate. (A) Multiple newly mated queens are known to often attempt to found colonies in groups. However, soon after the first workers eclose all but one queen is executed creating a monogyne colony. (B) Multiple monogyne colonies could fuse and maintain more than one queen, but monogyne colonies are territorial and there is no evidence that fusion between queenright monogyne S. invicta colonies occurs. (C) Newly mated queens could be adopted by queenright monogyne colonies; however, conspecific worker aggression from queenright colonies extends to newly mated queens. In laboratory experiments 100% of introduced newly mated queens were executed by monogyne workers. (D) Male and female sexuals within a monogyne colony could mate within the nest (intranidal mating), thus creating a polygyne colony. There is no evidence that supports intranidal mating, and it is known that queens in United States polygyne colonies are unrelated. None of the above appear to explain polygyne S. *invicta* in the United States.

We suggest a mechanism for the initiation of polygyny that involves adoption of NMQs into queenless worker groups. Insight into the mechanism of polygyne formation in fire ants came from the discovery that highly aggressive and territorial fire ant workers from monogyne colonies quickly become less aggressive toward conspecifics after they lose their queen. In addition, these queenless worker groups readily fuse to form new colony groups that contain several matrilines and patrilines, as are found in polygyne colonies. Queenless polygyne and monogyne colony workers, as well as fused monogyne worker groups adopt newly mated fire ant queens (normally executed by workers with queens). We propose that the probability for development of polygyne populations increases with the formation of population-wide queenless worker groups. Chemical treatment of soil, controlled burns, and fire ant toxic baits could lead to the population-wide formation of queenless worker groups, providing the conditions for the development of a polygyne population. We used toxic baits to test our working hypothesis that formation of multiple queenless worker groups would promote the development of polygyny within a monogyne population in the field. Of five replicate monogyne field plots treated with bait toxicant (hydramethylnon) four developed patches of polygyne colonies within 54 weeks after treatment, whereas all associated controls maintained their monogyne status. This is the first time that an association has been demonstrated between bait treatment and the development of polygyne fire ant colonies.

Fire ant queen pheromones: the when and where of secretion

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Abstract: Fire ant queens produce several different pheromones that modify the behavior and physiology of the different colony members in a profound way. A better understanding of these pheromones and their mode of action may provide new means to help manage fire ant populations. Many previous studies have demonstrated that fire ant queens produce both releaser and primer pheromones. In this study, I used established bioassays to investigate two queen pheromones: 1) the releaser pheromone underlying queen recognition and attraction by workers; and 2) the primer pheromone that prevents virgin queens from deaiating (shedding their wings). Previous work has shown the poison sac to be the source of both of these pheromones. Virgin queens were allowed to dealate and begin to lay eggs by placing them in small queenless colony fragments removed from the inhibitory influence of the queen. After dealation, virgin queens were tested at intervals of 24 hours to determine when the two pheromone systems switched on in relation to the onset of egg-laying. Virgin queens became significantly more attractive than controls 2 days after shedding their wings, at which time many (30%) of the virgin queens began to lay eggs. The primer pheromone was detectable only 24 hours later at 3-days post-dealation. Thus, both the releaser and primer pheromones are secreted at the initiation of egg-laying. In another series of experiments, I found that surgical removal of the poison sac from queens and virgin queen dealates did not cause the loss of either the releaser or primer effects, suggesting that in addition to the poison sac there is another source of these pheromones. Testing extracts of different body parts in the attraction bioassay indicated a source of activity in the head Research is currently underway to identify the unknown glandular sources of these pheromones.

Adoption of Newly Mated Queens: What are the consequences for Control?

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Abstract:

We previously demonstrated that territorial and highly aggressive fire ant workers become non-aggressive toward conspecific workers and newly mated queens (NMQs) soon after the loss of the colony queen. The decrease in aggressivity leads to requeening of the colony through adoption of NMQs. Here we attempt to answer the question - how does the adoption of NMQs relate to the control of fire ants?

The formation of queenless worker groups in the field can occur in many ways. For example, the colony queen can die from (A) natural causes, (B) mechanical disturbances, (C) chemical treatment of soil, and/or (D) bait insecticide treatment. Isolated polygyne colonies exhibit heightened conspecific aggression, similar to that found for monogyne colonies. For this reason we hypothesize that isolated polygyne colonies will have a low probability to develop into a polygyne population. If multiple NMQs were adopted by queenless worker groups from situations A or B above, they would likely lead to an isolated polygyne colony and not a polygyne population. Situations C and D have the potential to create population-wide queenless worker groups and polygyne populations.

Bait formulated hydramethylnon was used for fire ant control throughout the 1980's to the present. The action of this insecticide is known to kill the queen but not all the workers. We demonstrated a correlation between the field treatments with hydramethylnon bait and the development of incipient polygyne populations. But this represents a worst case scenario where attempts to control a monogyne population results in development of a polygyne population. Treatment produced queenless worker groups may never have the opportunity to adopt NMQs or ultimately adopt only a single queen yielding a new monogyne colony. Newly mated queen adoption, regardless of the monogyne/polygyne end result will lead to faster than expected reinfestation. The usual explanation for reinfestation of treated field sites is that colonies on the edge of the treated area move in to fill the void and/or NMQs reinfest treated areas. We want to add the adoption of NMQs by queenless worker groups as another option to consider when explaining reinfestation. Future research will define the effects of Fenoxycarb, Avermectin, and the presence or absence of alates on newly mated queen adoption.

Modeling Fire Ant Range Expansion

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Vinson (1997) summarized range expansion scenarios for the red imported fire ant (Solenopsis invicta). The extent of range expansion depends upon the temperature limits set for colony mortality (Calabi and Porter 1989). We decided to use our existing colony growth model (Korzukhin et al. 1997) to assess the northern limits of fire ant expansion.

We applied our fire ant single colony growth model to different geographical locations in 12 southern states. The basic model is driven by daily soil temperature and answers the question: "can a colony reach maturity under a given set of average daily temperatures?" Our model:

1. Uses NOAA weather data from 2090 stations as input (Alabama-136, Arkansas-138, Florida-171, Georgia-135, Louisiana-144, Mississippi-117, North Carolina-176, Oklahoma-176, South Carolina-90, Tennessee-116, Texas-539, Virginia-152). To avoid the complexities connected with running 12 years of weather data over 2090 sites, we used average daily weather values of Airmin, Airmax and Precipitation for each Julian day.
2. Converts max-min air temperature to max-min soil temperature at 10 cm (Chang et al. 1994, Kluender et al. 1993). Additionally, Chang et al. (1994) found that "..the inclusion of soil water content did not improve the predictability of the developed models." We found that the inclusion of precipitation did not improve our model. With respect to temperature. precipitation worked exactly like a random number variable.
3. Calculates colony growth (Porter 1988). mortality (Calabi and Porter 1989) and alate production (flight activity is governed by daily precipitation and peak abundance of alates occurs in May and June) (Bass 1974. Markin et al. 1974, Tschinkel 1993, Wojcik, unpublished data).
4. Assumes that production of one reproductive (alate) begins another colony. to sustain a fire ant population.
5. Estimates colony growth at each NOAA weather station.
6. Maps fire ant reproductive success by weather station (on an approximate 20 mile grid).

Figure 1 shows the gross northern limit of fire ant range expansion for southern states predicted by the model. Note that the southern Appalachian Mountains escape fire ants to some degree. This line fits closely the -18°C line theorized by Hung and Vinson (1978) and supported by Francke et al. (1986). Also, because precipitation is needed to trigger alate flights, **and** precipitation is sparse in the southwestern U.S., our model probably does a poor job estimating range expansion to the southwest. We suspect that our model will work in locations where irrigation is common because irrigated tracts probably mimic areas where precipitation is ample.

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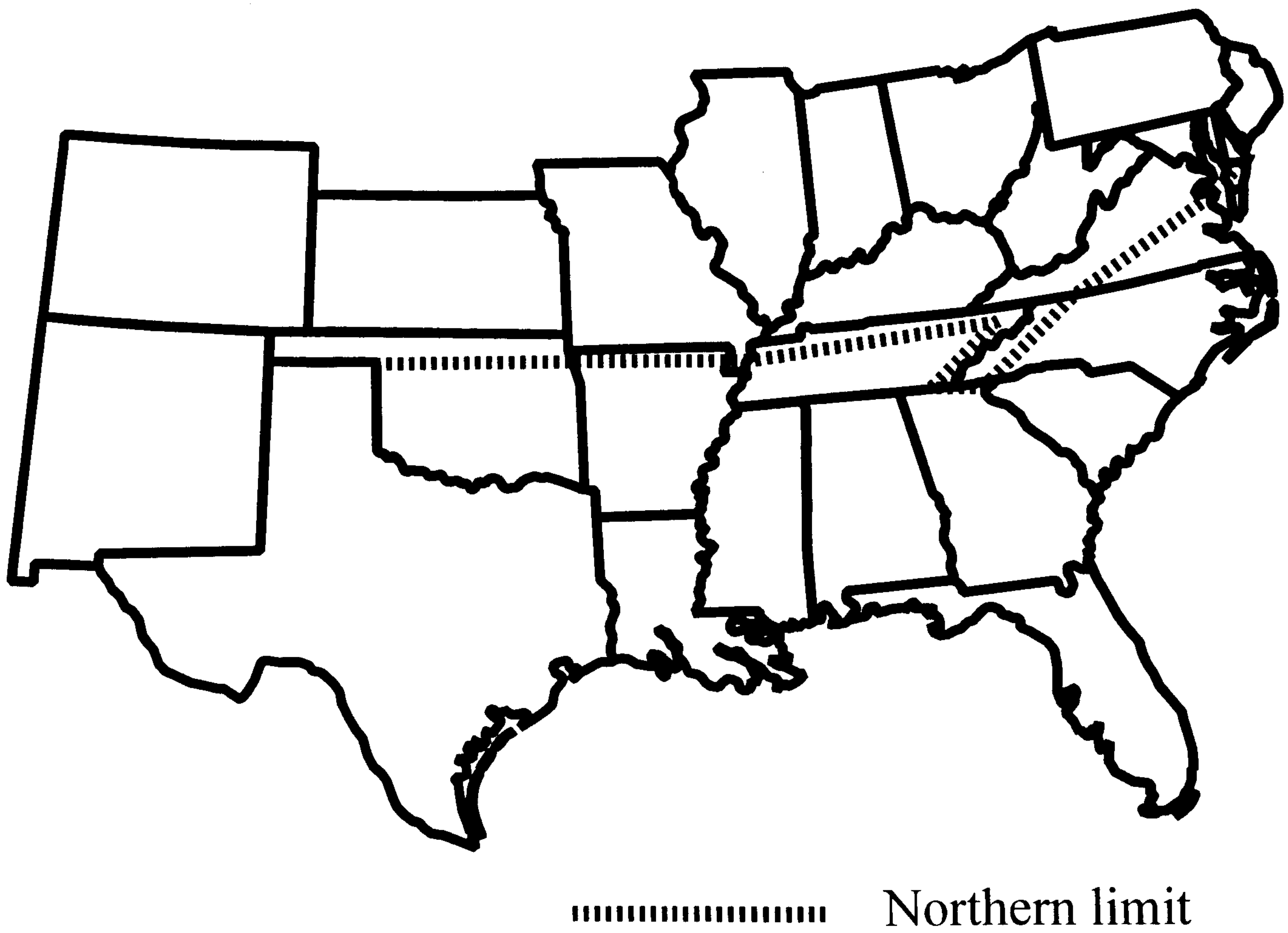
Fig. 1. Range expansion estimated by model



A Concept to Develop an IFA Temperature Suitability Model Using Remote Sensing and GIS

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INTRODUCTION

The Red Imported Fire Ant, *Solenopsis invicta* Buren, (IFA), was introduced to Alabama in **1930** and has spread rapidly to adjoining states. It has now colonized **13** states, as well as Puerto Rico. IFA were first reported in Union County, Arkansas, just north of the Louisiana border in **1959.** They currently inhabit at least 28 of Arkansas' **75** counties.

It has been estimated that fire ant damage currently results in an annual **$35** million loss to residents of southern Arkansas (Thompson, unpublished data). The influx of IFA populations into the northern part of the state is of great concern, not only to residents, but also to numerous large companies which transport products nationwide from northwest Arkansas (WalMart, Tyson, J.B. Hunt, etc). A model to hazard-rate fire ant range expansion is needed to assess the biological realities of colonization and the social and economic consequences that follow fire ant occupation. Reliable forecasting of colonization success in areas threatened by IFA infestation will allow policy makers and administrators to plan, fund, and implement regulatory activities.

OBJECTIVES

Development of this hazard-rating system includes three kinds of models. First, a colony biophysical model; second, a spatial model of physical features including vegetation cover and soil temperatures; and third, a socio-economic model of human and economic endeavors. Models will use accessible data bases and spatial statistics to develop spatial models utilizing physical features, vegetation classification, and human and economic parameters. Discriminate analysis will be used to assess what parameters best predict spatial patterns. The resulting spatial model will eventually be linked to the fire ant biophysical model to generate a hazard rating from 0 to 1. The proposed hazard rating model (equation 1) will be a function of soil temperature, (st), biophysical requirements, (b), social, (s), and economic parameters, (e). The interrelationships of these components is the focus of this study.

$$\text{Fire Ant Hazard Rating} = f\,(st,\ b,\ s,\ e) \tag{1}$$

PROCEDURES

Project description

This manuscript will elaborate on the concept of developing a spatial model using physiographic features to predict temperature suitability for IFA colonization. Numerous attempts have been made to model IFA range expansion (Pimm and Bartell, 1980; Cokendolpher and Phillips, 1989; Stoker, et al., 1993; Killion and Grant, 1995; Weih and Thompson, 1997). However, these models consider only homogenous habitats and do not take into account the dynamics of topography, soil temperature, and vegetation, all important parameters in potential colony initiation and development. Spatial statistics techniques in a Geographic Information Systems (GIS) framework will be used to incorporate heterogeneity into the landscape model. Examples of potentially useful landscape physical features include slope, aspect, soils, vegetation cover, and disturbance. GIS has the capabilities needed to spatially analyze these databases and determine their respective effect on soil temperatures. Remote sensing will be used to obtain satellite data which will reflect surface temperatures. These will then be modeled into predicted soil temperatures based on GIS layers and thermal probe base data. The resulting temperature suitability model will be used to produce a map surface which defines predicted IFA range expansion at a 1.1 km (121 ha; 300 ac) minimum mapping area.

Building a soil temperature data base

Soil temperature is one of the most relevant factors to colony survival. Unfortunately, soil temperature data is not currently being collected by any weather service station in Arkansas. Thus, an important part of generating the temperature contour map will be examining soil temperature as it is influenced by elevation, aspect, slope, and vegetation cover. To develop a soil temperature database, an experiment is being established in various ecologic regions in northern Arkansas to measure temperature at 5 and 50 cm depths. The 5 cm depth reflects temperatures needed by the fire ant to grow and develop. The 50 cm depth reflects the depth fire ants are likely to move to avoid killing winter temperatures. The experiment will involve two parts. Phase 1 will investigate the effect of elevation and aspect on soil temperatures in grazed pastures, a habitat highly suited to fire ant development. The experimental design includes 3 replicates, with measurement of temperature at 4 aspects (N, E, S, W) and 4 relative elevations (300, 600, 900, 1200 ft.). A thermocouple will be installed at the 50 cm soil depth at each temperature plot. Temperature at the thermocouple will be read monthly. Monthly measurements at 50 cm are adequate because temperatures will not vary much from month to month. Temperature measurements at the 5 cm depth and at 10 cm above the soil surface will be taken using electronic temperature sensors that record and store in memory readings at 1-hr intervals. Air temperature is needed to model the relationship between air and soil temperatures. Air temperatures will also be used to verify the air temperature model developed using satellite data. Soil temperature as a function of air temperature will then be used to develop a contour map for pasture environments.

Phase 2 of the soil temperature experiment, will involve setting up a replicated experiment in pine and hardwood forests to assess soil temperatures at 5 and 50 cm depths under a habitat of forest canopy or clearcut. Probes will be established at slopes of 0 and 10-20% on a south aspect. Temperatures will be measured as previously described.

All probe locations will be recorded using global positioning systems (GPS) equipment. These data will be imported into a GIS layer and surface temperatures collected by the probes will be used to verify satellite derived surface temperatures predicted for that specific location. Soil temperatures collected by some probes will be used to develop the function to convert surface temperatures to soil temperatures. Other probes will provide data to test the model.

Modeling surface temperatures using remote sensing

Since there are many thousands of climatological stations across the United States, one would think that accurate surface temperatures would be easily obtainable. However, the distribution of these stations across Arkansas is quite limited. There are 138 stations across the whole state that collect daily temperatures and of these, only seven are Automated Surface Observation System stations that collect hourly temperature data. Because temperature is one of the most important variables in determining successful colonization of fire ants, it is critical that an accurate temperature map be developed. This can be achieved by using satellite remote sensing to generate a pixel by pixel (1.1 km square; 121 ha; 300 ac) data base of vegetation surface temperatures across northern Arkansas. Thus, surface temperatures can be assessed at a finer spatial resolution than by using the climatological network alone. This finer resolution will result in a more heterogenous dataset.

Advanced Very High Resolution Radiometer (AVHRR) images will be obtained from the Satellite Active Archive of the National Oceanic and Atmospheric Administration (NOAA). Multiple dates of imagery will be used. This technology is carried on NOAA's Polar Orbiting Environmental Satellites (POES) and consists of a wide-band, five channel scanner that senses the visible, near-infrared, and thermal infrared portions of the electromagnetic spectrum. Each pass of the satellite provides a 2399 km (1491 mi) wide swath. The Local Area Coverage format has a ground resolution of approximately 1.1 km from an orbit altitude of 833 km (517 mi). Before the image can be used, it must be registered and georeferenced to known points on the ground. This will be done using ERDAS Imagine software. Channels 4 and 5 collect spectral reflectance in thermal infrared bands. Channel 4 is from 10.3 to 11.3 microns and channel 5 is 11.4 to 12.4 microns. In order to retrieve accurate values of land surface temperatures from satellite thermal infrared data, it is first necessary to calibrate the raw data. Some factors which contribute to the difficulty of converting to surface temperature are atmospheric water vapor, viewing angle, and channel surface emissivity which changes with vegetation cover. Several investigators have developed algorithms to perform the necessary corrections (Sobrino, et al., 1991; Kerr, et al., 1992; Vazquez, et al., 1997). Before the data can be used in a quantitative manner, it must be calibrated based on appropriate procedures. Temperatures will then be mapped and verified using data from national weather stations and strategically placed ground probes as ground truth locations.

Climate data will be obtained from the National Climatic Data Center (NCDC). Approximately 20-30 stations will be selected from Arkansas and surrounding states. Station locations, day, and time will be matched with cloud free AVHRR images. Latitude and longitude coordinates will be recorded for each station so that respective temperatures can be imported into GIS layers and compared with computed temperatures from the thermal infrared sensors of the POES satellites. Testing may reveal that other GIS data layers (such as slope, aspect, vegetation cover, or elevation) need to be incorporated into the model which predicts accurate surface temperatures from the AVHRR images. Equation 2 defines predicted air temperature as a function of AVHRR band 4, **(b4)**, AVHRR band 5, **(b5)**, physiographic features, **(p)**, and vegetation cover, **(v)**.

$$\textbf{Air temperature} = \mathbf{f\ (b4,\ b5,\ p,\ v)} \tag{2}$$

Modeling soil temperatures

Once the surface temperature layer is defined, the satellite information will be merged with the soil temperature data to produce a heterogeneous pattern of possible fire ant habitation sites displayed in the form of a contour map. Equation 3 illustrates that soil temperature is conceptually a function of air temperature, **(at)**, as it is derived in equation 2.

$$\textbf{Soil temperature} = \mathbf{f\ (at)} \tag{3}$$

The validity of this model will be tested using additional soil probe data from locations, dates, and times consistent with the image collection.

CONCLUSION

Reliable forecasting of colonization success in areas threatened by IFA infestation will allow Arkansas policy makers and administrators to plan, fund, and implement regulatory activities. This may save millions of dollars in economic loses attributed to fire ants. If the fire ant hazard-rating system can be successfully developed in Arkansas, such states as California, North Carolina, Oklahoma, Tennessee and Virginia may be interested in using parallel systems to estimate where fire ants might cause significant problems to their citizens and natural resources. This research will also develop innovative methodologies that will be useful to other states, the federal government, and private enterprises.

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Research Update on *Solenopsis (Labauchena) daguerrei*, a Parasitic Ant of Imported Fire Ants

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Summary

Solenopsis (Labauchena) daguerrei (Santschi) is a parasitic ant of imported fire ants. This parasite produces no worker caste, and is totally reliant on its host colony for its care. Having no worker caste, only reproductive males and females represent this species. *S. daguerrei* will attach, or yolk, themselves to queens of the black and red imported fire ants, *Solenopsis richteri* and *S. invicta*, respectively, and divert resources fiom the host queen(s). In addition, the host colony also feeds and maintains the brood of *S. daguerrei*. Thus, *S. daguerrei* is a potential stress factor of fire ant colonies. This parasite is found in South America, and is being held in quarantine in the United States at the Center for Medical, Agricultural, and Veterinary Entomology in Gainesville, Florida.

Field studies and observations of the impact of *S. daguerrei* on *S. richteri* were conducted in sites located in Buenos Aires Province, Argentina. Mean percent parasitism from pastures located at 21 sites (San Eladio, Argentina) was 5.1% (range: 1.2 to 23.7%; total 2,580 S. *richteri* colonies examined). Mound densities were 33% less in sites with S. *daguerrei*, where there were 161 ± 14 (± std. err.) moundsha, while parasite-free sites had 239 ± 15 moundsha. The number of *S. richteri* queens per colony was 47 % less in parasitized colonies (parasitized = 2.9 ± 0.5, range: 1 - 40 versus non-parasitized = 5.5 ± 2.0, range: 1 - 180). In addition, both female and male *S. richteri* alates were observed in parasitized colonies. This is in contrast to reports by Silveira-Guido et al. (1973) who did not observe alates in parasitized *S. richteri* colonies.

There was a 3:1 female to male sex ratio for S. *daguerrei* fiom excavated *S. richteri* colonies. However, colony was variation high where females in a colony ranged from 4.6 to 100%, and males ranged from 0 to 95.3%. Female alates were observed in field colonies in all months except Sept., Oct., and Nov., which is the spring in Argentina. Male alates were not observed in July through Nov. (winter-spring). *S. daguerrei* queens were observed in colonies throughout the year. The monthly presence of alates and queens in quarantine colonies corresponded to the field observations in Argentina. Alate flights by *S. daguerrei* were induced in quarantine by drenching and misting fire ant mounds located in buckets of soil. *S. daguerrei* were weak fliers, and mating was observed on the mound surface as well as in nest cells. Mating also occurred in the absence of flights.

S. daguerrei was not found in any of the non-fire ant colonies examined in Argentina from Dec. 1996 to March 1998. The following ant colonies were examined 92

Pheidole, 1 *Brachymyrmex*, 21 *Acromyrmex*, 1 *Linepithema*, 25 *Camponotus*, and 2 *Neivamyrmex*. The colonies that were examined were from areas where 4.7% (145 of 3,092) of the *S. richteri* colonies were infested by *S. daguerrei*. In quarantine, attempts to transfer *S. daguerrei* from *S. richteri* to *S. invicta* colonies have not been successful. As a result, attempts will now be made to develop methods of transfer from *S. richteri* to *S. richteri* before transfers across species are made.

Acknowledgement

Support for studies on *S. daguerrei* is provided through Dr. Lynne Thompson, University of Arkansas - Monticello.

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Pseudacteon/Solenopsis Interaction: Comparative Studies of Phorid Fly Oviposition Behavior and Responses to Attack by Worker Fire Ants

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Although phorid flies from the genus Pseudacteon directly affect mortality of worker fire ants, it is their indirect influence on ecological interactions with other ant species that makes them excellent prospects as biological control agents for the imported fire ant. The degree to which each of the phorid fly species impedes foraging and defense behavior of fire ant workers is a key indicator of this indirect effect. Therefore, this study focuses on comparing oviposition/attack behaviors of several Pseudacteon species, as well as subsequent responses by host ants to each fly species. The data presented are preliminary and qualitative, but they clearly indicate that adequate variation exists to predict differential potential among Pseudacteon species with respect to usefulness in biological control of the fire ant.

A good biological control agent must: be host specific, have mass-rearing potential, be able to cope with local climes, and, most importantly, be detrimental to its host. The research presented herein focuses on determining which of the phorid fly species would be the most detrimental to its host.

Actual rate of parasitism by phorid flies on fire ants is minimal, making it unlikely that the death rate in itself will suppress fire ant populations (Morrison et al. 1997). Rather, the key component lies in the effect the flies have on foraging and defense behavior. This has been established in several publications (Feener 1981, Orr et al. 1995, and Porter et al. 1995). Phorid flies effect foraging and defense behavior in two ways. First, ants in the vicinity of an attacking fly retreat into the nest or underneath nearby cover. If there is no cover, as in laboratory situations, the ants clump. Second, attacked ants become "frozen" in one of several postures. The function of these behaviors is not yet understood, but we hope to determine this with further research. These postures are relevant to the present study because while fire ants are "frozen" they cannot forage or defend resources. Thus their ability to compete with other species of ants is compromised.

One species of phorid fly, Pseudacteon *tricuspis*, is successfully being reared in two labs (Sanford Porter's of the USDA/ARS in Gainesville, FL and our own at Brackenridge Field Laboratories in Austin). Still, we are interested in rearing and releasing more than one phorid fly species for two reasons. First, within the North American distribution of the RIFA, there is tremendous variation in climates. Second, the various phorid fly species differ in time of day that they attack, what worker ant activity elicits attack (i.e., foraging trails versus disturbed mounds), and what size workers they prefer to attack. Our research seeks to identify phorid fly species with foraging ecologies that differ from that of *P. tricuspis*. The combined effect of more than one species will produce a more pronounced biological control effect.

To determine which phorid fly species elicit the greatest response from workers of the RIFA, we have begun testing of the following two hypotheses:

1) Attack behavior differs among species of *Pseudacteon* phorid flies.
2) Some species of phorid flies suppress foraging and defense behavior more effectively.

We completed observations in the laboratory of single female phorid flies attacking worker ants in glass-covered plastic trays. Worker ants were from more than 20 colonies. Phorid species for which observations have been completed include South American (*P. litoralis*, *P. tricuspis*, *P. obtusus*, *P. curvatus*, *P. nudicornis, P. comatus,* and *P. borgmeiri*) and North American species (*P. browni, and P. crawfordi*). We have the greatest experience observing attacking females of *P. tricuspis* and used this species as our benchmark species, against which all others were compared.

When phorid flies attack fire ants, the fly contacts the ant, presumably laying an egg in the thorax of the ant. An ant that is thus contacted by the attacking fly is referred to as the primary ant. Primary ants were immediately attractive to other ants, which approached the primary ant, antennate, and groom her. Ants that tended to the distressed primary ant were referred to as secondary ants. During observations of each of the species of phorid flies, we ranked three behaviors relative to the behavior of females of *P. tricuspis*: attack rate, primary ant response, and secondary ant response. All three of these behaviors are important measures of the degree to which a phorid fly species will have on the foraging and defense behavior of fire ants.

All three behaviors were ranked on a scale of one to five, five being the most intense rate/response (Figure 1). Attack rate of *P. tricuspis*, was scored as four stars. Primary ant response was ranked based on period of time the primary ant spent in the frozen posture. Primary ant response to *P. tricuspis* attack was scored as four stars. Quantitative data taken show that primary ants stay frozen for an average of approximately 6 min. Secondary ant response scores were based on how many ants responded to the primary ant and the length of time that secondary ants tended the primary ant.

Comparison among species of phorid flies shows clear differences among species with regard to all three behaviors observed. Furthermore, some phorid fly species do seem to have a greater effect on the foraging and defense behavior of fire ants. Clearly, some phorid fly species will be more promising biological control agents to pursue. Preliminary quantitative data have thus far confirmed the qualitative findings presented here.

Scale: ★ → ★★★★★

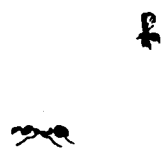
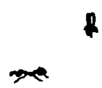
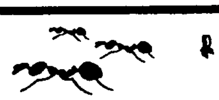
Pseudacteon sp (*n*, ex.)	Fly and Ant Sizes	Attack Rate	Primary Ant Response	Secondary Ant Response
borgmeieri (20, Argentina)		★★★★	★★★★★	★★★★
litoralis (20, Brazil)		★★★★	★★★★	★★★★
tricuspis (>100 Brazil)		★★★★	★★★★	★★★
browni (10, N. Am)		★★★	★★★★★	★★★★
crawfordi (10, N. Am.)		★★★	★	—
obtusus (5, Braz. and Arg.)		★★	★★	★★
curvatus (> 100, Braz. and Arg.)		★★★	★★★	★★
nudicornis (15, Argentina)		★★★	★★★★★	★★★★
comatus (5, Argentina)		★★★	★★	★

Figure 1. Comparison of the ethology of nine species of phorid flies and the fire ants they attacked. There were seven South American species of flies, which attacked the RIFA, *Solenopsis invicta*; and two North American species, which attacked the native fire ant, *Solenopsis geminata*. The first column shows the species of phorid fly, the approximate number of flies we have observed, and the location from which the fly came. The second column shows the relative sizes of the flies and the size of workers they preferred to attack. The last three columns show the relative scores each species of fly rated for attack rate, primary ant response, and secondary ant response, respectively. The benchmark species was *Pseudacteon tricuspis* (third species down in the column). See text for definitions.

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DIRECT AND INDIRECT EFFECTS OF PARASITOID PHORID FLIES

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The magnitudes of the direct and indirect effects of native Texas phorid flies (*Pseudacteon browni*, *P. crawfordi*, *P. spatulatus*, and *P.* sp. A) on native Texas fire ants (*Solenopsis geminuta*) were quantified. The direct effect of the parasitoid flies on host mortality was measured by subsampling workers from field colonies and counting the number of infected workers. Parasitism rates were found to be very low (< 3.0 %).

The indirect effect of phorid fly parasitoids results from the fact that phorids induce behavioral responses in their hosts which alter the outcome of interspecific competitive interactions. When phorids were present at baits, worker ant recruitment decreased, and most workers that remained at baits assumed a motionless, curled defensive posture. Phorids are only active diurnally, and observational experiments revealed that *S. geminata* obtained 50% less food by day than by night. Experimental manipulations which excluded phorids from foraging workers by day and allowed phorid activity after dark confirmed that the presence of phorids can affect resource retrieval rates and thus influence the exploitative component of interspecific competition.

The affect of phorids on the interference component of interspecific competition was measured by recording the outcome of contests at baits near the territorial boundary of *S. geminata* with its aggressive competitor, *S. invicta.* (The phorids native to the U.S. do not parasitize *S. invicta.*) The presence of phorids had no significant effect, however, on the outcome of these contests.

Thus the main effect of *Pseudacteon* phorid flies on *Solenopsis* fire ants in this system is an indirect one, behaviorally mediated through exploitative competition. If South American *Pseudacteon* species introduced to the United States as biocontrol agents interact with North American populations of *S. invicta* in a similar manner, biocontrol success will depend critically on: (1) the relative proportion of above-ground foraging, (2) the relative proportion of nocturnal foraging, and (3) the abundance of competing species which are able to usurp food resources abandoned by fire ants in the presence of phorid flies.

Field Release of Phorid Flies for Fire Ant Biocontrol

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Use of ABAMECTIN to Control Red Imported Fire Ants

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ABAMECTIN

- Abamectin Fire Ant Bait Development
- Mode of Action
- Product Description & Use Patterns
- RIFA Efficacy Summary
- Abamectin Fire Ant Products Available

NOVARTIS 2

Development of Abamectin

- Soil actinomycete fermentation product (*Streptomyces avermitilis)*
- Macrocyclic lactone in the Avermectin class (abamectin, emamectin benzoate and ivermectin)
- Composition: Avermectin $B1_a$ (≥80%) and Avermectin $B1_b$ (≤20%)
- Fire ant bait development initiated in 1980 by Merck & Co.

NOVARTIS 3

Abamectin - Mode of Action

- Acts as a GABA agonist in insects
- Against RIFA
  - Moderately toxic against worker ants
  - Sterilizes or kills the queen
    - Found to inhibit reproduction of queens at concentrations as low as 0.0025% (Lofgren & Williams, 1982)
  - Effects on queens are irreversible when applied at label rate

NOVARTIS 4

Abamectin - Mode of Action

- Reproductive Effects on RIFA
  - Tissue & Cell Abnormalities in Queens
    - Hypertrophy of epithelium around ovarioles
    - Pycnosis of nurse-cell nuclei
  - Very Few Eggs Produced
  - Egg size Reduced, lack yolk
  - Effects on neurosecretory cells
- Reproductive effects reversible only at rates of 0.00025% or lower

NOVARTIS 5

ABAMECTIN - Product Description

- Active ingredient: 0.011% avermectin B_1
- Formulated on PGD Bait (pregelled defatted corn grit)
- Trade Names

Trade Names	Company
VARSITY	Novartis
CLINCH	Novartis
ASCEND	Whitmire Microgen
RAID Fire Ant Bait	S. C. Johnson

NOVARTIS 6

1

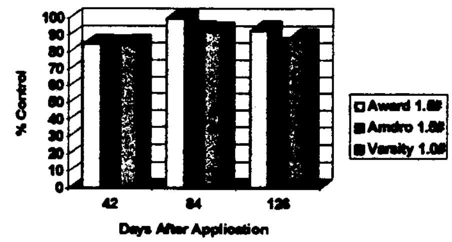
ABAMECTIN - Use Patterns

- Broadcast Application
  - 1 lb. ABAMECTIN Bait per acre (50 mg ai/A)
  - Ground Application Equipment
- Individual Mound Treatment
  - 5 to 7 Tablespoons ABAMECTIN Bait
  - Spread uniformly around 2 ft radius

NOVARTIS 7

% Control of Red Imported Fire Ants

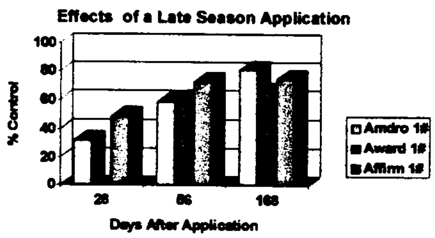
H. Collins-USDA, MS (applied 5/20/87) CI4397468 PASTURE



NOVARTIS 8

% Control of Red Imported Fire Ants

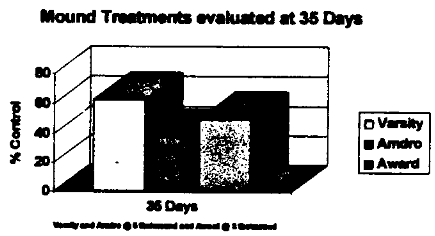
H. Collins-USDA, MS (applied 10/15/87) CI4397470 PASTURE



NOVARTIS 9

% Control of Hybrid Fire Ants

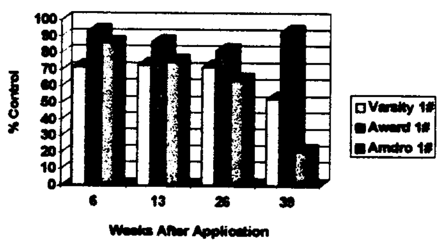
S. Diffie-Univ. of GA (applied 6/6/85) CI450333 PASTURE



NOVARTIS 10

Multi-Queen Colony Trials with Fire Ant Baits

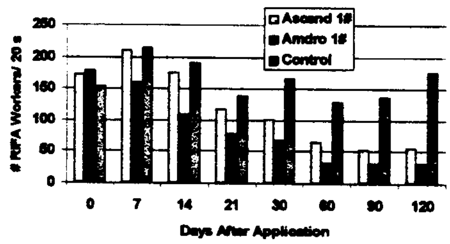
USDA, FL (applied 9/86)



NOVARTIS 11

Mean No. RIFA Workers / 20 sec

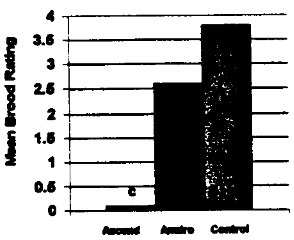
J. Vogt & A. Appel, Auburn (Fall 1996) Whitmire Data Pecan Orchard



NOVARTIS 12

Effect of Abamectin on RIFA Brood

J. Vogt & A. Appel, Auburn 1996-97 — Whitmire Data, Pecan Orchard

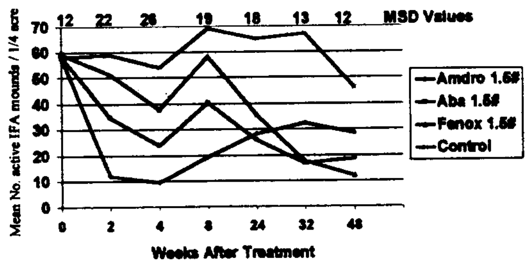


- Brood Ratings at 30 DAT
  - 0=No brood
  - 1=<1" diam
  - 2=1 to 2" diam
  - 3= 2 to 3" diam
  - 4= 3 to 4" diam
  - 5= >4" diam
  - Means tested w/ t-test

NOVARTIS 13

IFA Control - Mound Counts

B. Drees & C. Barr, TX, (Applied 9/10 & 9/12/96) Pasture

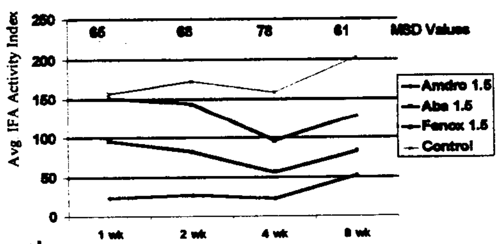


NOVARTIS 14

IFA Control - Mound Activity

B. Drees & C. Barr, TX (Applied 9/10 & 9/12/96) Pasture

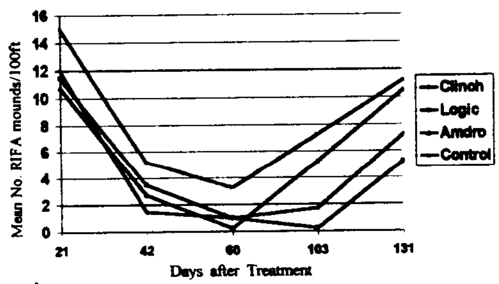
Ratings: 3 = > 1,000 ants, 2 = 100 - 1000 ants, 1 = < 100 ants. Index=Sum(Rating*#mounds)



NOVARTIS 15

Red Imported Fire Ant Control In Citrus Beds

(Stansly & Fulcher 1997, Univ. Florida)



NOVARTIS — All treatments applied at 1.0 lb./A. 16

ABAMECTIN - Summary of Efficacy Data

- ABAMECTIN is highly effective in reducing numbers of RIFA mounds
- Speed of Activity: Amdro>=Abamectin>Fenoxycarb
- Longevity of Control: Abamectin=Fenoxycarb>=Amdro

NOVARTIS 17

Abamectin Fire Ant Bait Marketing Channels

Market Sector	Product	Company	Comments
Turf (professional)	VARSITY	Novartis	Available Fall 1998
Agricultural	CLINCH	Novartis	Bearing citrus
PCO	ASCEND	Whitmire MicroGen	PCO Use
Consumer	RAID fire ant bait	S. C. Johnson	OTC, Mass Markets

NOVARTIS 18

Parasitization of the Red Imported Fire Ant by *Caenocholax fenyesi*: Host size selection and the utilization of alates

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Summary

Caenocholax fenyesi Pierce Strepsiptera: Myrmecolacidae) parasitizes all castes of the red imported fire ant, *Solenopsis invicta* Buren, with the possible exception of the queen. The first instar larvae of C. *fenyesi* are responsible for selecting a new host. This stage must parasitize either the larval or pupal stage of S. *invicta* for it to successfilly complete its development. Almost immediately after burrowing into its future host, the first-instar molts and becomes a secondary larvae, which is a maggot-like stage. The strepsipteran then goes through at least two more molts before entering the pupal stage. By the time *C. fenyesi* enters the pupal stage, S. *invicta* is an adult. Depending upon which larva was chosen, the strepsipteran will be in either a worker, female alate, or male alate. The strepsipteran emerges from its host as a free-living adult male (females develop in the bush cricket *Hapithus agitator* Uhler). However, strepsipterans appear to be unable to emerge from alate hosts.

Our study suggests that *C. fenyesi* selects larger hosts to parasitize. We collected over 200 male strepsipterans and their corresponding host ants. In doing so, we found a relationship where the size of the emerging male strepsipteran is directly correlated to the size of its host. We also compared the host size of parasitized ants with the host size of the normal size distribution of their respective colonies. Ow data suggested that strepsipterans are parasitizing hosts that are significantly larger than the general population from their respective colonies. Therefore, C. *fenyesi* may be choosing larger hosts.

This study found that both male and female alates were parasitized at a higher rate than worker castes. This data corresponds with the above portion of the study that suggested that strepsipterans selected larger hosts. If this is true, then *C. fenyesi* may be having a greater impact on its host population than if it were selecting hosts randomly. However, our study is not yet complete and more collections are needed to support this conclusion.

Ecological suppression of fire ants by dipteran parasitoids: A perspective from work based at Brackenridge Field Laboratory

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Abstract: Describes a program to understand the comparative biologies of fire ants and phorid fly parasitoids aimed at eliminating the pest status of IFA in Texas

BAIT STUDIES ON FIRE ANTS & FIRE ANT TOXICANTS ON TEXAS CAVE CRICKETS & FISH-BAIT CRICKETS

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Abstract: Field studies using attractant non-toxic baits demonstrated that fire ants foraged intensively in and around the caves. These studies also demonstrated that cave crickets emerged from the caves and foraged widely around the caves. The objectives were to assess susceptibility of endangered cave species to impact by fire ant and fire ant management practices and chemicals. As it was not feasible or legal to test endangered cave species, cave crickets, a scavenger species important in the cave food chain were used as a surrogate. The cave crickets live and reproduce in the caves and forage for food outside of the caves. Cave crickets were tested in two ways. Cave crickets were fed on specific numbers of bait granules to determine the risk Amdro bait poses to the crickets. Cave crickets were fed on Amdro poisoned lire ant cadavers, to determine if the movement of toxicants via scavengers into the cave systems was possible. Additional tests were conducted using fish-bait crickets as surrogates for cave crickets. Amdro granules were toxic to cave crickets and fish-bait crickets feeding on bait granules. Amdro poisoned fire ant cadavers did not pose a threat to the cave crickets. The ants could easily be controlled without affecting the cave crickets by restricting fire ant treatments to morning hours, thus allowing sufficient time for the lire ants to forage for and retrieve all of the bait particles before the cave crickets emerge from the caves during the night.

Impact of native ants on fire ant colony foundation

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ABSTRACT: The impact of native ant species on Solenopsis *invicta* colonies in the laboratory and greenhouse are being studied. Laboratory techniques similar to the ones developed by Bhatkar (1988) were used, where an established native ant colony were given access to founding fire ant colonies with varying numbers of workers. The behavioral interaction between native ant species and fire ants, and time until the fire ant colony disruption, were recorded. The studies used the native species, M. minimum, Pheidole dentata, *Forelius* sp., and *Tetramorium bicarinatum* versus founding fire ant colonies of various sizes. The results suggest that M. minimum can invade and destroy founding fire ant colonies within 7 days when worker number is in the range of 50 to 60. The other native ant species such as Pheidole dentata, *Forelius* sp., Tetramorium bicarinatum differentially attack on fire ant founding colonies with various numbers of workers and brood. However, the time taken for these latter species to destroy fire ant colonies with up to 50-60 workers was observed to be longer than the former two species. All the species were found to predate on dead fire ant workers and the brood. Similar studies are being conducted in the greenhouse arenas with the species studied in the laboratory. The experimental set up in the greenhouse simulates the field conditions and the experiments are currently underway.

Aerial Application of Fenoxycarb for Fire Ant Control on a Quail Plantation: An Interim Report

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ABSTRACT

Increased numbers of imported fire ant colonies have been implicated in the decline in quail populations on a south Georgia quail plantation. Aerial application of fenoxycarb on the 4000 acre plantation reduced the number of active fire ant colonies, as well as the number of foraging IFAs, as compared to untreated plots. After treatments, the number of non-formicid insects collected in sweeps was higher in the treated plots than in the untreated plots, suggesting an increased availability of food for quail with the elimination of imported fire ants.

INTRODUCTION

Imported fire ants may affect quail populations by attacking young quail (Giuliano et al. 1996) and/or by competing for food resources (Allen et al. 1995). The foraging ability of IFAs can cause the demise of other ants (Vargo and Porter 1989), other arthropods (Porter and Savignano 1990), and other animals through competition and predation (Allen 1994). After establishment of colonies in an area, the competitiveness of imported fire ants prevents native ants from reentering the area.

MATERIALS AND METHODS

Location and Design

- Mossy Dell Quail Plantation (4000 acres) in Lee County, Georgia.
- 500 acres of Mossy Dell were divided into 10 plots containing 4 subplots each. Each subplot contained 2 sampling sites.
- Fenoxycarb was applied to five plots at the rate of 1.5 lbs per acre on 23 June, 1996; 29 October, 1996; and 30 April, 1997.
- Bait application was accomplished using Transland spreader fitted with a seeding plate and mounted on a turbine Ag Cat.

Sampling

- Counts of active IFA colonies were taken annually at 2 subplots per plot.
- Foraging data were taken biweekly by placing a test tube containing 2 g of canned tuna at each sampling site. The tubes were left for one hour, stoppered with collected ants inside, and returned to the lab for specimen identification and counting. The number of ants in

each tube was determined and indexed according to the following scale: 0=no ants, 1=1-10 ants, 2=11-50, 3=51-100, 4=101-150, 5=151-200, 6=201-250, 7=251-300.

- Sweep net samples--25 per subplot--were taken monthly during winter months and biweekly during the remainder of the year to quantify non-target arthropod populations.

RESULTS AND DISCUSSION

There has been a notable reduction in the number of foraging fire ants and a concomitant increase in the number of foraging non-target ants (Figure 1) in the treated plots since the initiation of this program. In the untreated plots, foraging activity of IFAs followed a seasonal (fall/winter) reduction in numbers but recovered the following year, while the non-target ants remained low throughout the trial (Figure 1). The number of active IFA colonies decreased significantly in the treated compared to untreated plots (Figure 3). The number of non-formicid insects collected in the sweeps followed a seasonal trend but was numerically higher in the treated plots than in the untreated plots suggesting a recovery of species in the absence of IFAs (Figure 2).

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(nonIFA) in relation to fenoxycarb treatment. Ants collected in tuna bait stations (n=40 per treatment). Vertical lines indicate treatment dates.

IFA/Quail Study
Mossy Dell Plantation, Lee County, GA

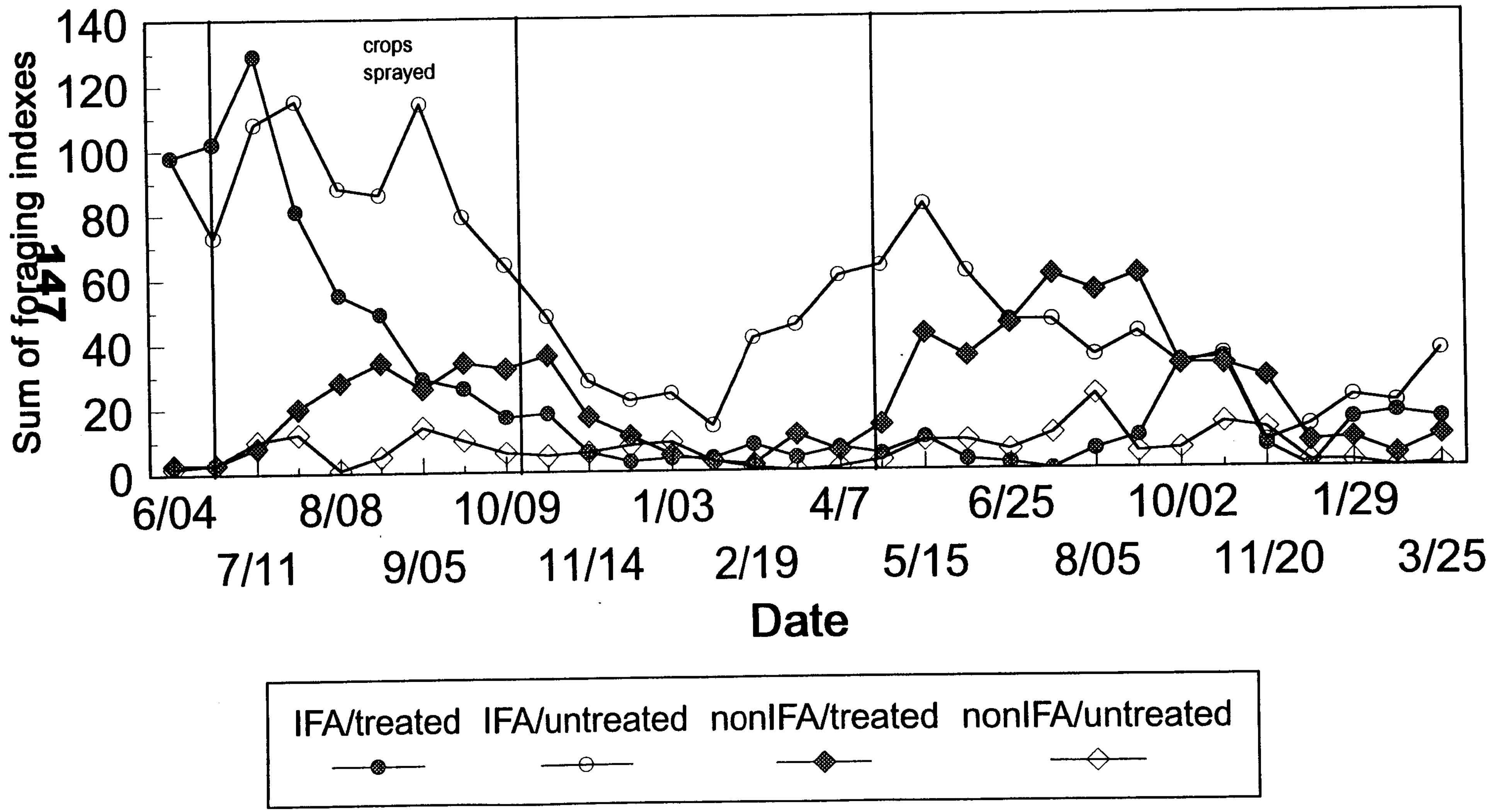


Figure 2. Sums of foraging indexes for imported fire ants (IFA) and number of arthropods collected (per 200 sweeps) in relation to fenoxycarb treatment (treated plots). Vertical lines indicate treatment dates.

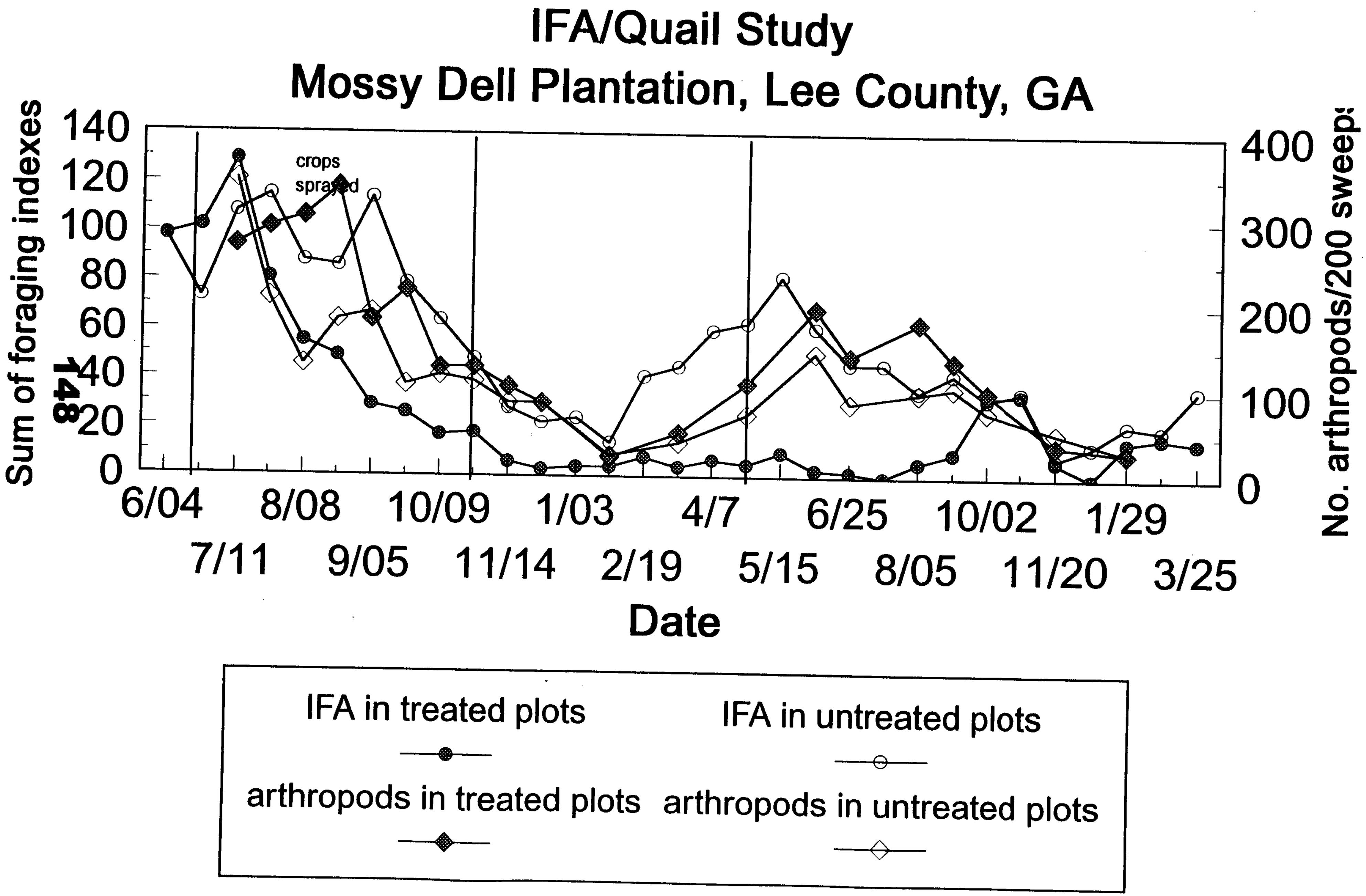
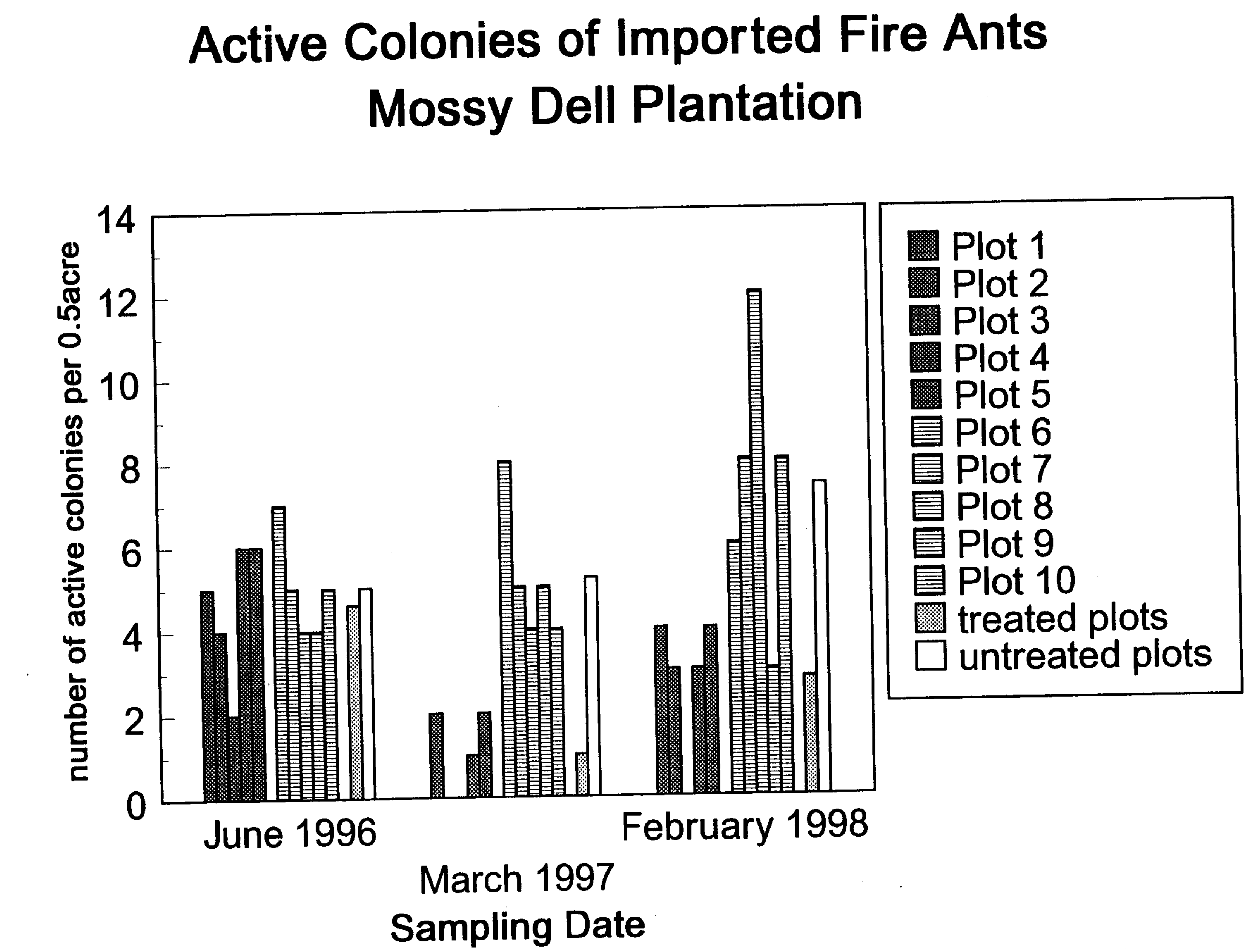


Figure 3. Number of active imported fire ant colonies per 0.25 acre. Counts in June 1996 were prior to treatment. Plots 1-5 were treated with fenoxycarb, and plots 6-10 were untreated. Mean of treated and untreated plots are represented by green and yellow bars, respectively.



TITLE: Control of Red Imported Fire Ant (Solenopsis *invicta)* With Spinosad
Author(s): T. Craig Blewett, Raymond B. Cooper
Address: Dow **AgroSciences** LLC, Indianapolis, IN 46268

Abstract:

Spinosad was formulated as 0.01% or 0.02% granular baits and as a liquid drench for evaluation as a control agent for Red Imported Fire Ant (Solenopsis invicta) at several locations in the Southeastern US. Applications consisted of **2-**6 tablespoons of bait per mound, 1.5 and 3 **lbs** per acre broadcast, or **½ -** 2 gallon mound drenches of 25 **-** 400 ppm solutions of Conserve™ SC, all to well established mounds. Commercial treatments applied for comparison were **Amdro®** bait, **Dursban®** granules and Dursban drenches. Ant morbidity was generally observed within 24 hours after treatment with dead ants being piled outside mounds in 2448 hours. Spinosad-based treatments provided approximately 50% control of all mounds by 3 days after treatment and **>90%** control by 14-21 days after treatment. Spinosad treatments compared favorably to commercial insecticide treatments.

Introduction

The Red Imported Fire Ant (RIFA), Solenopsis *invicta*, has become an important economic pest and public health threat in the U.S. since its introduction in the 1930's. It has spread along the Gulf Coast and now infests an area from Texas to North Carolina. The ant inhabits urban yards as well as agricultural and recreational areas. It is particularly noxious due to its aggressive behavior, attacking and injecting a venom with its sting, resulting in intense burning and itching. Attempts at eradication have failed and **current** control measures are generally limited to individual mound and small broadcast treatments. Control products include drenches, surface-applied contact toxicants and baits.

Spinosad is a combination of spinosyns which are a naturally derived group of molecules from the actinomycete, *Saccharopolyspora* spinosa. Spinosad is active as both a contact and ingestion toxicant but is generally much more active via oral administration. It has a unique mode of action acting at the nicotinic acetylcholine receptor site. It is particularly efficacious against Lepidopterous larvae but has shown activity across nine orders of insects, including Hymenoptera. Spinosad was evaluated in 1997 to determine if activity was present to warrant commercial development as a **RIFA** control product.

Materials and Methods

For individual mound and broadcast trials spinosad was formulated into 0.01% and 0.02% baits using defatted corn grit as the carrier. Conserve SC was diluted to provide the appropriate spinosad concentrations for drench trials.

Trials were conducted in South Carolina, Georgia and Florida in areas with well established mounds and actively foraging ants (Figure 1). Spinosad treatments consisted of 2-6 tablespoons of bait spread evenly around the mound (Figure 2), 1.5 and 3 lbs per acre broadcast or ½ to 2 gallon mound drenches of 25 - 400 ppm solutions. Broadcast applications were made with a hand held rotary applicator. Comparison treatments were Amdro and Dursban 1/2G granules for mound and broadcast bait treatments; Dursban 1/2G followed by a 1 gallon water application over the granules for drench trials. All mound treatment trials used one mound per replication with 10 replications per treatment. Broadcast applications were made to ¼ acre plots with three replications per treatment.

Fire ant control was assessed at various intervals using a 0 or 100 rating with a zero assigned if any live ants were observed after slightly disturbing the mound. A 100, indicating complete colony kill, was assigned if no live ants were found after significantly disturbing the mound. Data from trials were combined where appropriate and Locally Weighted Regression (Loess) lines plotted.

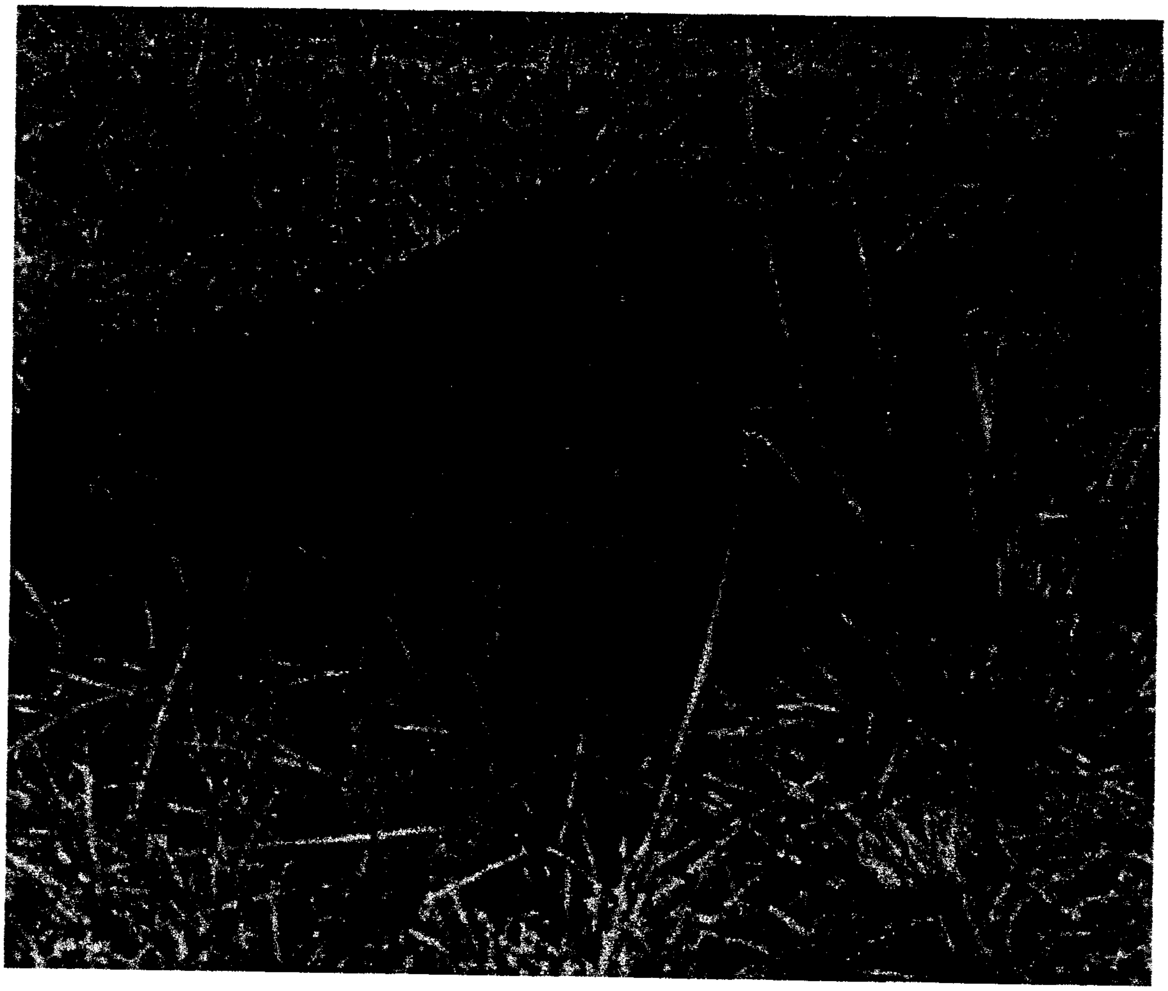


Figure 1. Well established RIFA mound typical of those included in trials.

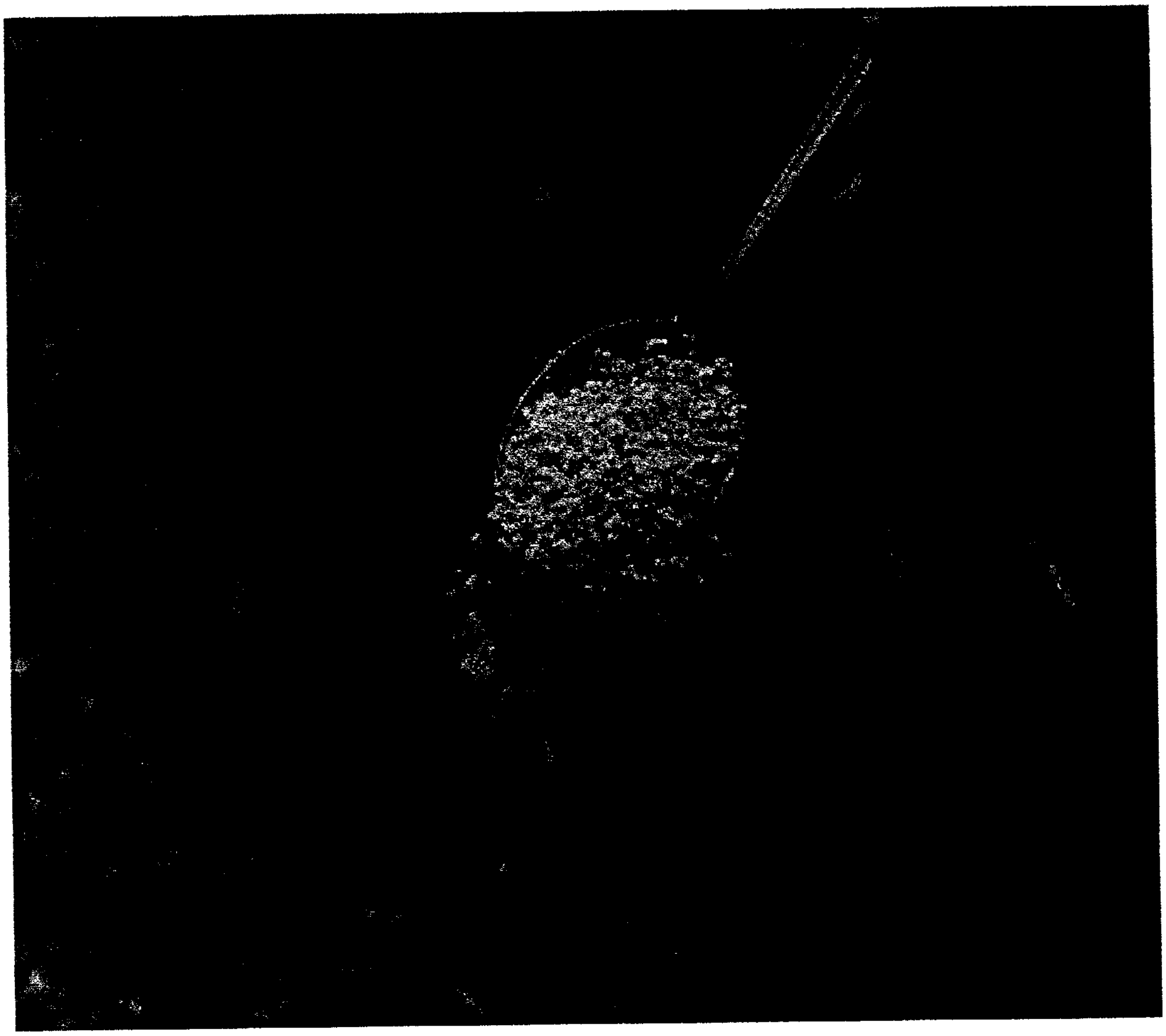
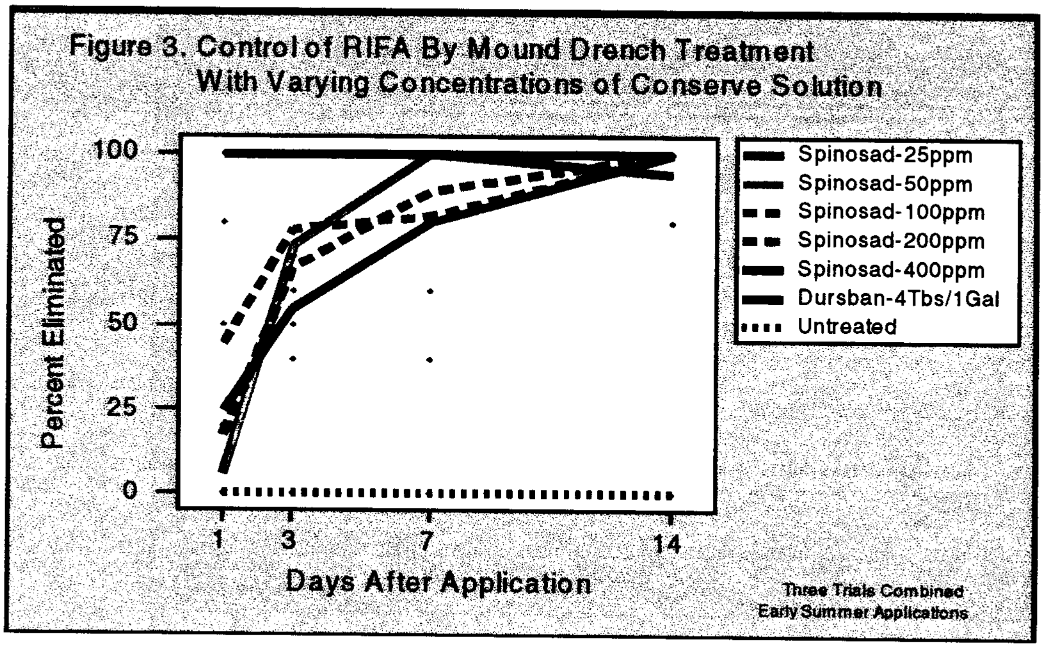


Figure 2. Application of bait treatments to individual RIFA mounds.

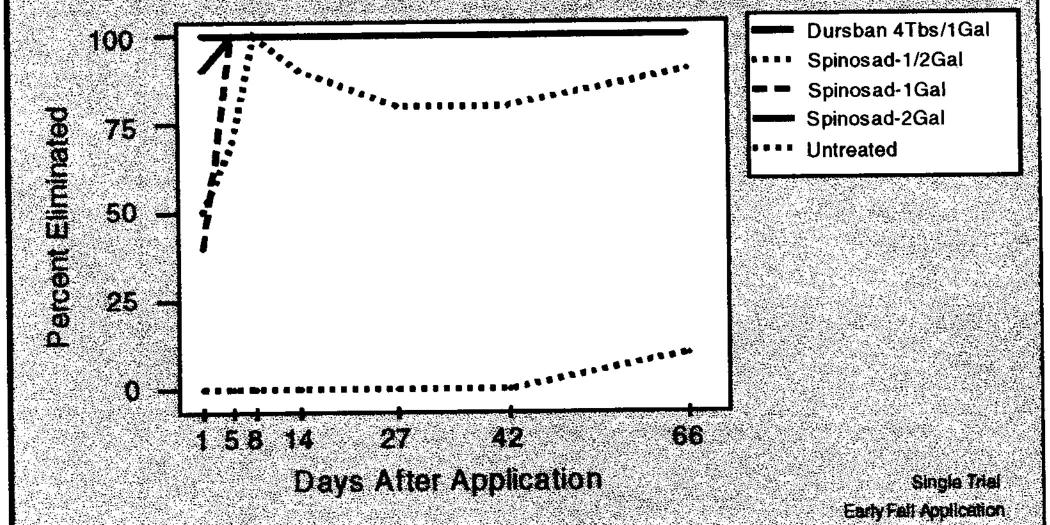
Results and Discussion

Spinosad provided very effective control of RIFA when applied as a drench, individual mound treatment or as a broadcast application. Complete cessation of activity was observed in approximately 20% of the colonies after one day with 1 gallon drench applications and all treatments showed greater than 75% of the colonies eliminated by 7 days (Figure 3). Control with spinosad treatments was slower than with Dursban, however all treatments were near 100% control at 14 days after treatment (DAT). No clear dose response was observed, but the 100 ppm treatments appeared to have quicker initial activity while the 25 and 50 ppm treatments were the first to reach near 100% elimination. A shallow dose response with spinosad treatments has been observed with other insect species (M. Tolley, L. Larson, pers. comm.). Although spinosad has contact activity, the much slower activity compared to Dursban may indicate that ingestion is the principle route of exposure for spinosad with drench treatments.



The volume of treatment solution may account for a greater initial response than the concentration used. In a trial varying the volume of a 100 ppm solution, a 2 gallon application showed activity almost as fast as Dursban (Figure 4). By 5 DAT there was no difference between Dursban and spinosad treatments at 1 or 2 gallons per mound. The ½ gallon treatment appeared to completely control the colonies by 8 DAT but by 66 DAT approximately 10% of the colonies recovered. Higher drench volumes may allow the treatment solution to reach greater depths within the mound resulting in direct exposure to a larger percentage of the population.

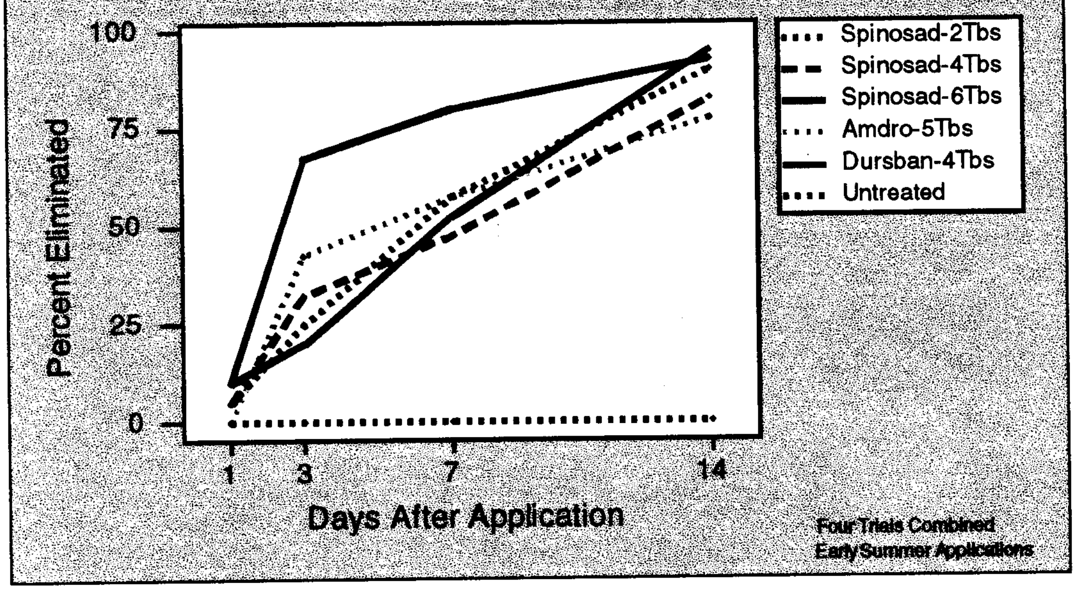
Figure 4. Control of RIFA By Mound Drench Treatment With Varying Volumes of 100 ppm Conserve Solution



Initial granular bait trials compared different application rates of a 0.02% spinosad bait with labeled rates of Amdro and Dursban 1/2G. Ants readily foraged both the spinosad and Amdro baits, completely removing the material from the soil surface in 30 to 120 minutes (Figure 5). Neither degree of control nor speed of kill were appreciably influenced by spinosad bait application rate (Figure 6). By 14 DAT there was little difference among treatments, with all providing better than 75% control. Amdro appeared to provide slightly faster control at 3 DAT but by 14 DAT gave lower overall control. The acute contact activity of Dursban compared to bait materials was apparent at 3 and 7 DAT. The quick action of a contact toxicant is often compromised if the colony is not quickly eliminated because the ants may abandon the treated mound and establish new "satellite" mounds within a few feet of the original mound. In these trials Dursban was not "watered in" which accounts for the slower activity and lower overall efficacy compared to the drench trials (Figures 3 and 4). The slower acting baits allow the toxicant to be disseminated throughout the colony with little or no repellency, increasing the potential for total control.

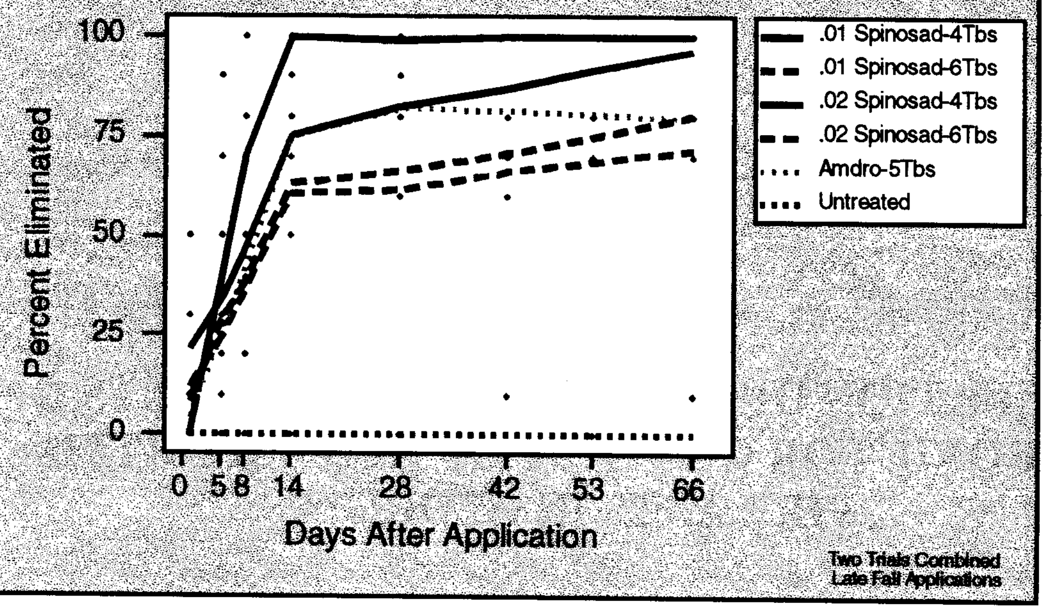
Figure 5. Foraging RIFA with bait granule.

Figure 6. Control of RIFA By Mound Treatment With Varying Rates of Spinosad Baits



A second set of individual mound bait trials was performed to determine if a 0.01% bait would be as efficacious as the originally formulated 0.02% bait. Two rates of both concentrations were applied and compared with Amdro. Results at 14 DAT were similar to the previous bait trial but there was wider separation between spinosad treatments out to 66 DAT (Figure 7). Additionally, the data sets were much more variable. At one location, 4" of rain fell within a few hours of application and could have influenced bait integrity and ant foraging. A preliminary observation showed the 4 tablespoon rate for both bait concentrations provided superior control compared to the 6 tablespoon rate. This observation will require further testing to interpret.

Figure 7. Control of RIFA By Mound Treatment With Varying Concentration and Rate of Spinosad Baits



In the mound applied trials Spinosad bait granules were actively foraged with no indication of repellency. The onset of toxicity was observed within 16-24 hours of treatment. Ants that were present on the mound or that emerged after slightly disturbing the mound were noticeably less aggressive and overall movement was erratic. Within 24 hours dead ants were being removed from nests treated with either spinosad or Amdro. Dead ants from the Amdro treatments were initially placed in structured piles at a distance from the mound openings. In the spinosad treatments the dead ants were not placed in the structured piles and were not removed any appreciable distance from mound openings. It appeared that spinosad was rapidly distributed throughout the worker class, causing behavioral changes.

Broadcast applications of 0.02% spinosad bait were made at 1.5 and 3 lbs per acre and Amdro was applied at 1.5 lbs per acre. There was a clear separation of spinosad treatments in speed of kill but by 66 DAT there was little difference between treatments in overall efficacy (Figure 8). The 3 lb rate of spinosad and 1.5 lb rate of Amdro gave similar control and speed of kill. These trials demonstrate that control of RIFA can be accomplished with very small amounts of active ingredient. The 1.5 lb rate of spinosad 0.02% bait is equivalent to 0.0003 lbs of active ingredient per acre.

EVALUATION OF BROADCAST TREATMENTS OF FIPRONIL FOR CONTROL OF RED IMPORTED FIRE ANTS IN GEORGIA

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ABSTRACT

The broadcast application of products for the control of imported fires can be more advantageous than individual mound treatments. Three rates of Exclude (fipronil 0.1%G) were applied to a grass parking area at the Georgia National Fairgrounds in Houston County, Georgia, in June 1997. An experimental fipronil bait, Dursban 50WP, and Amdro also were broadcast in this trial. The broadcast application of fipronil granules for the control of imported fire ants afforded over 96% control of the active colonies throughout the 30-week trial. All of the treatments significantly reduced the number of active colonies. Compared to the untreated control plots, all treatments significantly reduced the population index. From this trial, it is evident that the broadcast application of fipronil granules is an effective treatment for the control of imported fire ants.

INTRODUCTION

The red imported fire ant (*Solenopsis invicta*) can be found in all 159 counties in Georgia. Research and Extension faculty at The University of Georgia estimate that Georgians expend approximately $48.5 million dollars per year in order to meet fire ant quarantine requirements in nursery and sod production operations, to cover medical costs related to fire ant stings, to repair damage to equipment due to fire ant mounds, and to purchase materials to control imported fire ants.

Current management programs for the red imported fire ant rely upon repeated broadcast applications of fire ant baits with follow up treatment of surviving colonies using an individual mound treatment technique. Persistent use of this combination of control techniques provides long term control of fire ant populations in the treated areas.

Evaluation of new products and control techniques for fire ant management is an ongoing research program at The University of Georgia. This study evaluates the efficacy of a broadcast application of fipronil in suppressing fire ant populations when compared with broadcast applications of a contact insecticide (Dursban), a fire ant bait (Amdro), and an experimental bait (Exp 61443A).

METHODS

The test was conducted on the grounds of the Georgia National Fair in Houston County, Georgia. Twenty-one fire ant-infested 0.8-acre plots were marked off in the perimeter grass parking area surrounding the complex. Each plot contained a 0.25-acre circular sampling area. Prior to treatment, the number of active fire ant colonies, as well as the fire ant population index was documented for each subplot. Treatments were applied on June 17, 1997 using broadcast applicator equipment. Evaluations (see Figures 1 & 2 for materials and rates) were made by counting the number of active fire ant colonies in the 0.25-acre areas at 2, 4, 6, 8, 12, 16, and 30 weeks posttreatment. Population index ratings were made at each posttreatment evaluation using the Lofgren and Williams (1982) modified scale indexing scale. An LSD test ($P=0.05$) using SAS for Windows was used to analyze the data and separate the means.

RESULTS

A significant decrease in the number of active fire ant colonies occurred within two weeks following application of Amdro, Dursban, and Exclude (0.1% fipronil) at the 0.01875 lbs AI/A and 0.0250 lbs AI/A when compared to the untreated area (Table 1). By four weeks posttreatment, all treatments showed a significant decrease in the number of active fire ant colonies when compared to the untreated control. Fipronil-treated areas maintained greater than 96% decrease in fire ant colonies relative to pretreatment counts throughout the remainder of the evaluation period.

Table 1. The percent decrease in number of active fire ant colonies in areas treated with Amdro, Dursban, Exclude (fipronil 0.1%G) at three rates, an experimental bait, and the untreated control at 2, 4, 6, 8, 12, 16 and 30 weeks following application. (Houston County, Georgia, 1997)

TREATMENT	Number of Colonies PRETRT	% decrease in # colonies at indicated week posttreatment						
		2	4	6	8	12	16	30
Amdro (1.5 lbs/A)	32	81.25a	100a	96.87a	100a	93.7a	78.1a	87.5a
Dursban 50%WP (8 lbs AI/A)	29	89.65a	100a	96.50a	100a	100a	96.5a	100a
Exclude (0.0125 lbs AI/A)	26	57.69b	96.15a	100a	100a	96.1a	100a	96.1a
Exclude (0.01875 lbs AI/A)	27	92.59a	100a	100a	100a	100a	96.2a	100a
Exclude (0.0250 lbs AI/A)	28	85.70a	100a	100a	100a	96.4a	100a	100a
Exp 61443A (1.5 lbs /A)	27	37.03b	81.48a	88.88a	92.5a	92.5a	88.8a	85.2a
Control	28	3.37c	39.29b	+3.57b	39.29b	35.7b	25.0b	32.1b

The percent change in population index also indicates a significant decrease in the size of all treated colonies, with the exception of experimental bait 61443A, within two weeks of treatment (Figure 2). Four weeks following treatment, all products resulted in a significant decrease in the population index of fire ant colonies, and suppression of the fire ant population was maintained throughout the test period.

Figure 2. The percent change in population index of fire ant colonies in areas treated with Amdro, Dursban, Exclude (fipronil 0.1%G) at three rates, an experimental bait, and the untreated control at 2, 4, 6, 8, 12, 16 and 30 weeks following application. (Houston County, Georgia, 1997)

Treatment	**Pop. index pretrt**	**% change in population index at indicated weeks posttreatment**						
		2	**4**	**6**	**8**	**12**	**16**	**30**
Amdro (1.5 lbs/A)	435	89.88a	100a	97.7a	100a	98.1a	77.0b	96.5a
Dursban 50%WP (8 lbs AI/A)	370	92.70a	100a	98.6a	100a	100a	99.4a	100a
Exclude (0.0125 lbs AI/A)	400	81.50a	99.2a	100a	100a	99.2a	100a	99.2a
Exclude (0.01875 lbs AI/A)	370	95.13a	100a	100a	100a	100a	95.0a	100a
Exclude (0.0250 lbs AI/A)	360	89.72a	100a	100a	100a	99.4a	100a	100a
Exp 61443A (1.5 lbs/A)	380	35.26b	85.5a	86.8a	92.1a	94.7a	90.7a	93.4a
Control	485	29.89b	62.2b	15.4b	67.4b	74.8b	37.9c	85.1b

Exclusion of RIFA Colonies from Containerized Nursery Stock with Spin Out® in Combination with Pyrethroid Insecticides

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Animal and Plant Health Inspection Service
Plant Protection and Quarantine
Gulfport Plant Protection Station

Spin Out® root growth regulator technology was developed by Griffin Corporation to enhance the development of dense, compact root growth. The active ingredient is copper hydroxide contained in a latex matrix which is sprayed onto the interior of plastic nursery containers. Since 1995 we have been testing the hypothesis that the use of Spin Out technology, in combination with a fast acting pyrethroid insecticide such as bifenthrin or tefluthrin, might prevent RIFA invasion of nursery containers by creating a toxic barrier at or around the drain holes of the containers.

MATERIALS AND METHODS

TEST I

Standard trade one nursery containers (Lerio Corp., Mobile, AL) were treated by Griffin Corp. with various concentrations of bifenthrin (0.25. 0.5, and 1.0% AI) in Spin Out. Pots were shipped to the Gulfport laboratory and in October *1995, 25* pots of each bifenthrin concentration were filled with a standard potting media. After filling, pots were aged in a can yard under simulated nursery conditions, including overhead irrigation.

In January 1996, Griffin Corp. supplied us with the Spin Out product containing 0.1% tefluthrin. This mixture was applied with a paint brush to the interior of *25* pots. Pots were filled with media and aged as above.

Treatments were bioassayed monthly in the laboratory with field collected RIFA colonies. One container from each treatment was placed at the end of a 2' x 8' test arena. Sides of the test arena were coated with Fluon® to prevent escape. An untreated check container filled with the same media type was placed at the distal end of the arena. A field collected colony. complete with associated soil and nest tumulus, was then placed in the center of the arena. Overhead incandescent bulbs slowly desiccated the nest so that the ants were induced to migrate into the more moist containers. Therefore the colony had an equal opportunity to move into either a Spin Out treated pot or the untreated check pot. Pots were observed at 24 hour intervals for 7 days after introduction, and the estimated number of workers successfully invading each pot recorded.

TEST II
Griffin Corp. supplied Spin Out (a different formulation than used in Test I) containing bifenthrin (0.25, 0.5, and 1.0% AI by weight). Lerio Corp. provided the containers and applied the Spin Out/bifenthrin to the pots using automated equipment. The coating was applied to the pots at a rate approximately ⅓ the thickness of the preliminary trial, therefore the amount of insecticide available in each pot was also reduced by ⅓. This was done for improved pot stacking. The nurseries listed below provided media, space, and plants for the trial. Trials were set up by APHIS, Lerio and Griffin personnel.

NURSERY	LOCATION	1-gal Plants	3-gal Plants
Turkey Creek	Houston, TX	*Buxus microphylla*	*Buxus microphylla*
Grass Roots	Leesburg, FL	*Juniperus parsonnii*	*Ilex crenata*
Flowerwood	Loxley, AL	*Ilex crenata*	*Ilex crenata*
Windmill	Folsom, LA	*Ilex crenata*	*Ilex crenata*
Wight	Cairo, GA	*Juniperus horizontalis*	*Juniperus horizontalis*

Pots were transported to the Gulfport lab for bioassays as described above. Bioassays were performed at 1 and 3 months after potting, and quarterly thereafter.

RESULTS AND DISCUSSION

TEST I
All rates of bifenthrin in the Spin Out 100 technology successfully prevented RIFA infestation in nursery pots for 24 months (Fig. 1). Tefluthrin also successfully prevented RIFA infestation for 24 months, although a few workers (<30) were found under one tefluthrin treated pot at 11 months. The Spin Out 100 technology without an insecticide does

not exclude RIFA from containerized nursery stock.

TEST II
Preliminary results through 9 months show some movement of colonies (workers and brood) into pots treated with 0.25% and 0.5% bifenthrin (Fig. 2). During all evaluation periods, a few workers (<100) moved into or under some of the 3-gallon pots treated at 1.0%. Less than 50 ants moved into one of the 1.0% 1-gallon pots at 6 months after potting up.

DISCUSSION

Results from the first test initiated in 1995/1996, demonstrated the potential for Spin Out technology combined with a pyrethroid insecticide as quarantine treatment for containerized plants.

In the second test, initiated in 1997, it has become evident that with the Spin Out 300 formulation, the thinner coating of latex, and therefore less insecticide applied to the pots, has affected the efficacy of all treatments. It also appears that the larger pots (3-gallon), with proportionally larger drain holes, may require more insecticide per surface area.

New tests are being initiated this spring (1998) using increased rates of insecticide in the latex coating and different application techniques.

CONCLUSION

This technology, if shown to be cost-effective, could revolutionize the way nursery stock is certified for movement outside the IFA quarantined area. The requirement to blend granular insecticides into potting media or apply liquid drenches would be totally eliminated, and worker exposure greatly diminished.

Mention of trade names or proprietary products does not constitute an endorsement or recommendation for use by the U. S. Department of Agriculture.

Figure 1. Percent of Spin Out 100 + Insecticide Treated Pots Infested with IFA. Preliminary Trial at Gulfport, MS - initiated 1995 - 1996.

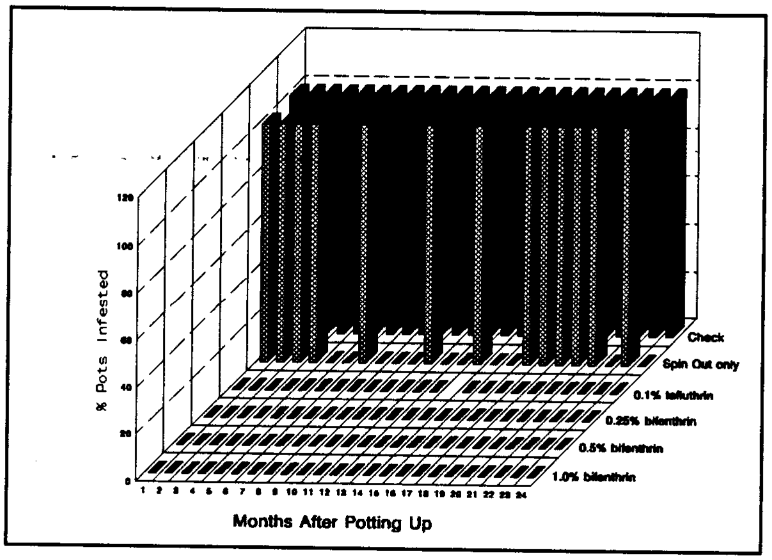
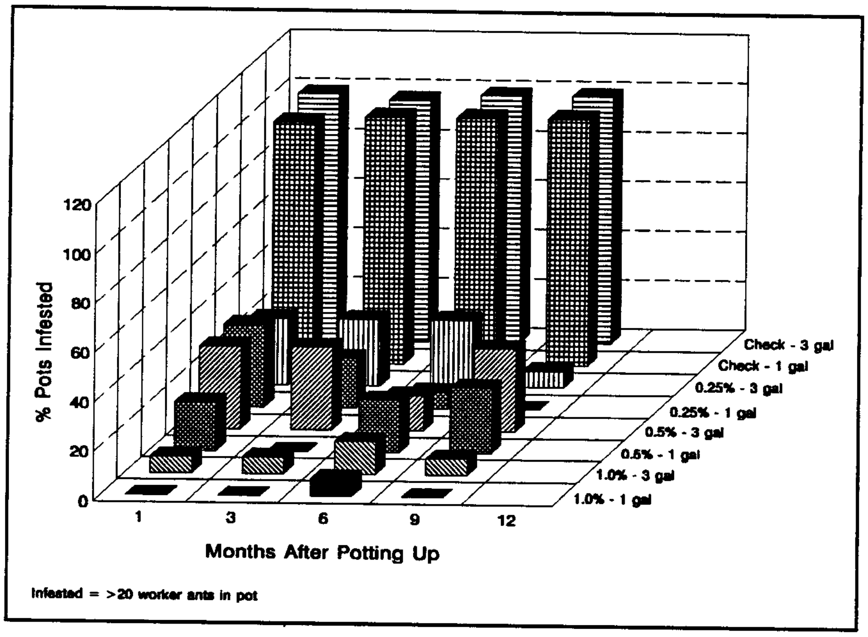


Figure 2. Percent of Spin Out 300 + Bifenthrin Treated Pots Infested with IFA. Multi-State Trial - initiated March 1997.



Fire Ant Control in a Hay Storage Area

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Imported fire ants had gotten so bad in Dan Wilson's hay storage yard that marketing his large hay bales was in jeopardy. He turned to the local county extension office for help in controlling this serious pest. An on-farm demonstration was established to show how an effective control program could drastically reduce fire ant populations and the resulting problems they cause. Prior to treatments, the mound counts were averaging over 240 mounds per acre on the farm near Stamps, Arkansas.

Pasture areas were treated with Amdro® broadcast at a rate of one pound per acre, using a HERD® broadcast seeder. Four weeks after this initial treatment, 90 percent of the mounds were inactive. The remaining mounds were treated with Orthene® 75S at one teaspoonful per mound (Table 1).

Both of these insecticides are currently labeled for use in pastures. The only restriction is that Orthene® cannot be applied to pastures and hay meadows lactating dairy cattle are present. ***(The Orthene label has since changed and pastures are NO LONGER on the label).***

This two-step program allowed Dan Wilson to get better grazing for his stocker cattle. Keeping ants out of hay bales is also a big plus when they are being moved or fed. "This program approach is the best system I've found to control fire ants" said Wilson. "Using this system, in the fall and again in the late spring, has resulted in an overall 90 plus percent control rate" said Wilson.

Table 1. Results of fire ant control demonstration at Dan Wilson Farm, Stamps, AR, treated June 17, 1997.

Treatment	% Active Mounds		%Inactive Mounds	
10 mounds per treatment	**14 DAT**	**28 DAT**	**14 DAT**	**28 DAT**
Outside Field Area				
Logic @ 1 #/acre	90	70	10	30
Pinpoint@ 1TBS/mound	0	0	100	100
Combat@ 2 tsp/mound	30	10	20	90
Inside Hay Storage Area				
Amdro@ 1 #/acre	80	10	20	90
Orthene 1 tsp/mound	30	10	70	90

DAT - days after treatment

Impact of the Imported Fire Ant on Biodiversity: Standardized Spatial Monitoring of Foraging Interactions

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The imported fire ant is an invasive species that has gradually spread to all or parts of 13 states. In recent years, tbe negative impact of this species on biodiversity has been recognized. We are developing methods to quantify the impact of fire ants on biodiversity using spatial analysis and precision targeting to develop and implement reduced-risk management strategies. Herein, we compare two bait/monitoring approaches, used in combination with spatial statistical analysis, to assess relative abundance and foraging profiles of several species of ants, setting the stage for future comparisons with the introduction of interventions to mitigate fire ant populations and enhancelsafeguard biodiversity of the ecosystem.

Previous studies on monitoring foraging of ants have utilized a wide variety of bait materials, including ground meat and honey (Wojcik 1994), tuna fish (Levins et al. 1973), peanut butter (Oi et al. 1994), beef or chicken sausage (Porter et al. 1992), creme sandwich cookies (Banks & Williams 1989), and cookie crumbs (Human & Gordon 1996). Each of these and other food materials that have been used to sample ants, have disadvantages that include availability, suitability, sanitation, and reproducibility. One of the major problems with most of these bait materials has been the range of species attracted to a specific bait. Species readily attracted to meat baits may not be as readily attracted to sweet baits, and visa versa. Meat and honey baits used simultaneously often produce dramatically different results (Glancey et al. 1976). Recently Vail et al. (1997) submitted a patent application for a attractant bait formulation which was attractive to a variety of ant species, including those attracted to meat or sugars. The logistical simplicity of the new attractant formulation appeared to present a large enough improvement over our standard hamburger bait to warrant testing.

Methods

Avon Park Studies: In conjunction with other studies on the effects of fire ants on endangered species, a comparison test was conducted using our standard hamburger (ground meat meatball) and the new multiple species ant attractant bait (MSAA), at the Avon Park Air Force Range at 9 AM on 30 May 1997. An 81-station grid was marked out with flags with approximately 6 feet (2 meters) between flags and approximately 6 feet between rows. After bait placement, GPS was used to verify actual locations of each station. The standard hamburger bait (314-inch diameter meatball on 1-inch square heavy aluminum foil) was placed on the soil surface. For pickup, the hamburger on the foil was scooped up into disposable plastic souffle cups and placed in a plastic bag for storage. The MSAA bait was previously pipetted onto a flattened cotton ball placed in a 50 mm diameter X 9 mm self-sealing petri dish (Gelman Sciences Inc.). For bait placement, the lid is removed from the petri dish and the base containing the saturated cotton is placed on the soil surface. For pickup, the lids were placed on the bottoms on the dish bottoms, sealing them against ant escape and the sealed numbered dishes returned to the original plastic shipping tray. Both baits were placed on the soil surface simultaneously by 2 participating researchers, about 1 foot (30 cm) on either side of the flag marker. The hamburger bait was always placed to the left of the flag while the MSAA was always placed to the right. Both baits were left in place for 1 hour, after which the samples were collected and refrigerated for return to the laboratory. Upon return to the laboratory, the samples in the dishes were frozen overnight to kill the ants, and processed for identification and counting.

Gainesville Studies: Additional tests were conducted at CMAVE, Gainesville, FL, on 16-17 July 1997. These tests were conducted to ensure that one bait did not interfere with the collections on the other bait. A 49-station grid, marked out with flags with 6 feet (approximately 2 meters) between flags and 6 feet between rows, was measured out in the grassy field behind the laboratories. The hamburger baits only were put out commencing at 9 AM, 16 July 1997, and the MSAA baits only were put out commencing at 9 AM 17 July 1997. The hamburger and MSAA baits were handled as previously described.

Spatial distributions of ants were assessed for each bait separately (Brenner et al. 1998). Contour maps were prepared for absolute counts and for "indicators" (presence/absence) using the default kriging algorithm in SURFER for Windows (ver. 6.04, Golden Software, Golden CO). Differences in estimated areas were determined by subtracting indicator "grids" of MSAA from those of HAMB (Brenner et al. 1998).

Results and Discussion

The species collected and distributions the differences in the indicators for each species of the Avon Park collections are given in Table 1. For red imported fire ants, Solenopsis invicta, the hamburger resulted in more specimens, but 71.2 % of the locations showed no differences between the occurrence of RIFA on the baits. The other species were collected in much fewer numbers on either bait. Both baits collected 6 species in common. Pheidole moerens and Solenopsis littoralis were only collected on hamburger bait. Pheidole morrisi and Dorymyrmex bureni were only collected on MSAA bait.
The species collected and the differences in the indicators for each species of the Gainesville collections is given in Table 2. For red imported fire ants, the hamburger resulted in more specimens, but 87.4 % of the locations showed no differences between the occurrence of RIFA on the baits. Pheidole dentata and Dorymyrmex bureni were only collected on MSAA bait.

Advantages of MSAA bait over hamburger bait

1. MSAA bait is attractive to ant species which feed on sugars (honeydew from Homoptera or extra floral nectaries). i.e. Dorymyrmex bureni and Solenopsis littoralis.

2: MSAA is attractive to species which feed on protein (meat). i.e. Solenopsis invicta and Pheidole species.

3. MSAA is attractive to species which are usually not attracted to protein baits. i.e. Cyphomyrmex rimosus and Odontomachus brunneus.

4. MSAA has distinct logistical advantages over hamburger.

A. Simpler laboratory preparation: pipetting vs rolling meatballs.

B. Freezing or refrigeration not as critical for transportation or holding in field prior to use.

C. Easier handling for placement on soil surface, meatballs easily roll off foil.

D. Petri dishes can be pre-numbered on the bottom, eliminating confusion in the field during sample distribution and collection.

E. Eliminates the need for the additional container (souffle cups) required by the meatball baits.

F. Cleaner samples. MSAA components are soluble in alcohol. Hamburger fat often adheres to the ants, creating difficulties in identification.

G. Easier pickup of MSAA baits. The white cotton or filter paper is easier to see than a meatball covered with ants.

H. More compact storage. The petri dishes are returned to the original plastic shipping trays, eliminating the bulky souffle cups in plastic bags, requiring less ice chest space in the field and less freezer space in the laboratory.

I. Eliminates sample loss since the petri dishes are placed in the original plastic shipping trays as they are collected and there is no danger of the dishes popping open during transport or freezing.

J. Speeds sample processing since the petri dishes can be placed in numerical order as they are collected.

K. Less waste: Aluminum foil not required; petri dishes require simple soak and wash, as compared to a grease removal from souffle cups when hamburger is used.

L. No sanitation problems: the distribution of meat baits requires washing of hands after handling meat baits and disposal of used meatballs.

Given the versatility and specificity of this method, we propose this method be used as a standard monitoring program. Standard methods of monitoring will be essential for measuring survival and impact of candidate biological control agents throughout the proposed release sites in the United States.

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Table 1. Differences in indicators of ant abundance on the two baits from the Avon Park study site. The differences in area occupied by each species and corresponding percentages are given. Hamburger bait and multiple species ant attractant bait (= MSAA bait) tested simultaneously.

Ant Species	Bait	Area (sq.	Percent
Solenopsis invicta	Hamburger Only	114.1	26.6
	MSAA Bait Only	9.6	2.2
	No Difference	304.8	71.2
Pheidole floridana	Hamburger Only	23.0	5.4
	MSAA Bait Only	0	0
	No Difference	405.6	94.6
Pheidole morrisi	Hamburger Only	0	0
	MSAA Bait Only	4.2	1.0
	No Difference	424.4	99.0
Pheidole moerens	Hamburger Only	3.7	0.9
	MSAA Bait Only	0	0
	No Difference	424.8	99.1
Solenopsis littoralis	Hamburger Only	20.6	4.8
	MSAA Bait Only	0	0
	No Difference	407.9	95.2
Dorymyrmex bureni	Hamburger Only	0	0
	MSAA Bait Only	1.2	0.3
	No Difference	427.4	99.7
Paratrechina faisonensis	Hamburger Only	26.5	6.2
	MSAA Bait Only	187.9	43.9
	No Difference	214.1	49.9
Cyphomyrmex rimosus	Hamburger Only	2.5	0.6
	MSAA Bait Only	3.6	0.8
	No Difference	422.4	98.6
Odontomachus	Hamburger Only	35.7	8.3
	MSAA Bait Only	10.0	2.3
	No Difference	382.8	89.4
Paratrechina concinna	Hamburger Only	2.0	0.5
	MSAA Bait Only	13.8	3.2
	No Difference	412.7	96.3

Table 2. Differences in indicators of ant abundance on the two baits from the Gainesville study site. The differences in area occupied by each species and corresponding percentages are given. Hamburger bait and multiple species ant attractant bait (= MSAA bait) tested on consecutive days.

Ant Species	Bait	Area (sq.	Percent
Solenopsis invicta	Hamburger Only	21.9	11.2
	MSAA Bait Only	2.8	1.4
	No Difference	171.3	87.4
Pheidole dentata	Hamburger Only	0	0
	MSAA Bait Only	14.5	7.4
	No Difference	181.5	92.6
Dorymyrmex bureni	Hamburger Only	0	0
	MSAA Bait Only	97.5	49.8
	No Difference	98.5	50.2

Imported fire ants and soil fertility implications.

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The imported fire ants (*Solenopsis* spp.) are notorious pests, but what are they doing to the soil? It is difficult to look out over a pasture full of their large mounds and imagine that they are not having a significant impact on the agroecosystem. The high densities of their large mounds combined with their high rates of mound relocation (Briano et al., 1995) mean that they will affect entire soil landscapes in a relatively short period of time. Their mounds are higher in the important plant nutrients phosphorus and potassium than the surrounding soil (e.g. Blust et al., 1982, Green et al., 1998), and this must be considered in its environmental context if long-term impacts on soil fertility are to be predicted.

The objective of this research was to evaluate the effects of imported fire ants (*S. richteri* and *S. invicta*) on soil phosphorus and potassium by a holistic approach—studying their effects on the composition, structure, and behavior of Mississippi soils.

Properties of mounds and underlying soils were compared with adjacent, undisturbed soils on eleven soils fiom eight Mississippi counties and six of the state's soil resource areas. Concentrations of phosphorus and potassium in the mounds were determined by the methods of Raspberry and Lancaster (1977) and Schollenberger and Simon (1945). Structural analyses included sectioning of resin-impregnated mounds, aggregate stability (Russell and Feng, 1947), and scanning electron (Lynn and Grossman, 1970) and light microscopy (Brewer, 1964). Soil moisture was determined gravimetrically, and temperatures were monitored with thermocouples (Scherba, 1962).

Mounds on all soils were higher in phosphorus and potassium than the upper 5 cm of the undisturbed soil (e.g. Table 1). This effect may be due to the importation of plant material into the mound by the ants. It represents the localized concentration of nutrients fiom the surrounding environment.

The soil below the mounds was also enriched in these elements, and this suggests that materials are being transported downward fiom the mound into the underlying soil, perhaps by leaching with water through the channel network. Microscopic studies revealed evidence of water transport. A lining of material observed along the wall of a channel 30 cm below a mound had the appearance of water deposited material.

The network of channels which fills and underlies the mound has a tremendous capacity for conducting water. Channels can occupy fiom 39 to 55% of the horizontal cross-sectional area of the mound, depending on the texture of the soil. The crust of the mound is often porous, relatively unstable in water (e.g. Table 2), and would not be expected to inhibit infiltration of water into the mound.

Mounds are often drier than the adjacent soil, especially in the upper part, but they also absorb water more rapidly during rainfall as water filters through the porous crust (e.g. Table 3). Infiltrated water penetrates more deeply into the soil beneath the mound than in undisturbed soil.

Increased water movement through and below the mound would induce greater leaching of constituents fiom the mound into the subsoil. The movement of constituents would be enhanced by the high temperatures in the mound, which would increase rates of nutrient release into mobile forms.

The mounds get hotter than adjacent soils, especially in the spring and summer months (e.g. Figure 1). These findings are consistent with those of Scherba (1962) and Pinson (1980). Temperatures at 5 cm depth in the mound were as much as 16.5 °C greater than the adjacent soil at the same depth. These increased temperatures would enhance rates of microbial decomposition of organic material and release of nutrient constituents into mobile forms.

The ants concentrate nutrients in the mounds from the surrounding landscape, cause the mobility of these nutrients to be enhanced, and expose the mobile nutrients to increased leaching deeper into the soil. It appears that the colony-mound-soil unit may function as a "funnel", draining nutrients from soil landscapes (Figure 2). Periodic mound relocation by colonies would enhance this effect by continually changing the locations of the funnels. The long-term impact of this process could be increased need for agricultural fertilizers and increased non-point source pollution in the southeastern United States.

Table 1. Mean phosphorus and potassium contents of imported fire ant mounds-underlying soils and undisturbed soils on a Vaiden silt loam in Northeastern Mississippi.

Depth (cm)	P (mg kg^{-1})		K (cmol kg^{-1})	
	Mound-Underlying Soil	Undisturbed Soil	Mound-Underlying Soil	Undisturbed Soil
31—29	68*		0.51*	
29—20	58*		0.46*	
20—10	54		0.45*	
10—0	60		0.51*	
0—5	45*	13	0.41	0.27
5—10	45*	5	0.36*	0.11
10—29	22*	5	0.16*	0.08
29—45	18*	6	0.14	0.09
45—65	9	4	0.11	0.10
65—85	5	4	0.10	0.11
85—100	3	3	0.11*	0.12

Note: Means within mound (above 0-5 cm depth) marked with asterisk significantly different (α=0.05) from 0—5 cm depth of undisturbed. Means below mound marked with asterisk significantly different (α=0.05) from equivalent depth in undisturbed.

Table 2. Mean weight of aggregates from crust and interior of mounds and control topsoil on a Grenada silt loam in Hinds Co., Mississippi remaining (from initial weight of 5 g) after oscillation in water for given time period.

Minutes	2	4	8	16	32
		---------- g ----------			
Crust	1.4 b	0.8 b	0.7 b	0.4 b	0.3 c
Mound	2.9 b	2.5ab	2.3ab	1.7 a	1.5 b
Control	3.9 a	3.5 a	3.2 a	2.5 a	1.9 a

abc= Means in same column and soil with same letter are not significantly different (α=0.05).

Table 3. Moisture content with depth in mound and adjacent soil on Grenada silt loam, on August 12, 1997, less than 1 day after 1.5 cm of rainfall, Falkner silt loam on August 13, 1997, less than 1 day after 1 cm of rainfall, and Falkner silt loam August 13, 1997, immediately after a rainfall event.

	Grenada		Falkner		Falkner	
	1 day after 1.5 cm		1 day after 1 cm		Immed. after rainfall	
Depth	Mound	Control	Mound	Control	Mound	Control
inches	---------------------%---------------------					
0-0.4	25.59	34.40	32.85	47.23	51.47	69.39
0.4-2	31.98	24.20	35.52	29.60	40.99	8.26
2-4	44.22	20.93	31.42	22.62	35.78	30.80
4-6	40.28	14.73	27.10	14.89	36.35	25.91

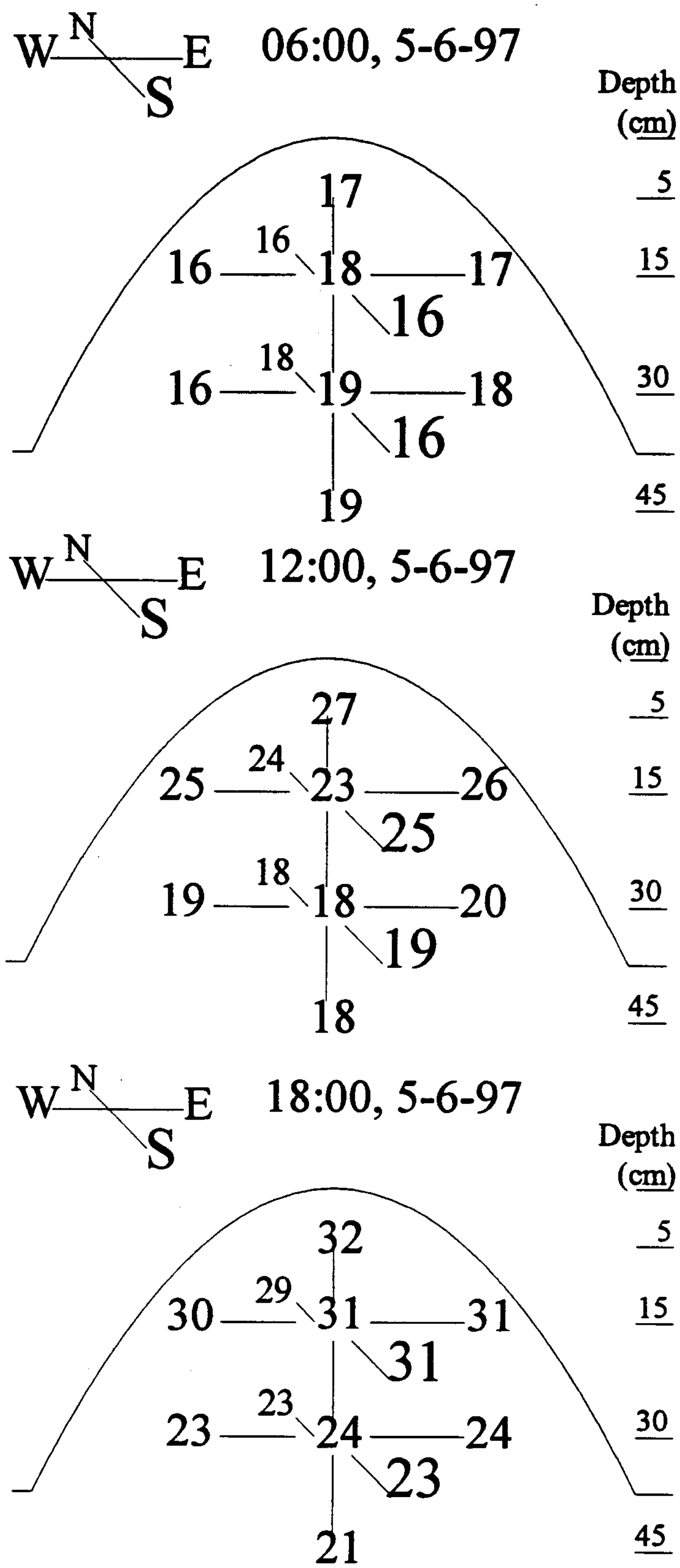

Figure 1. Temperature (°C) in a single *S. richteri* mound during May in northeastern MS on a Kipling soil.

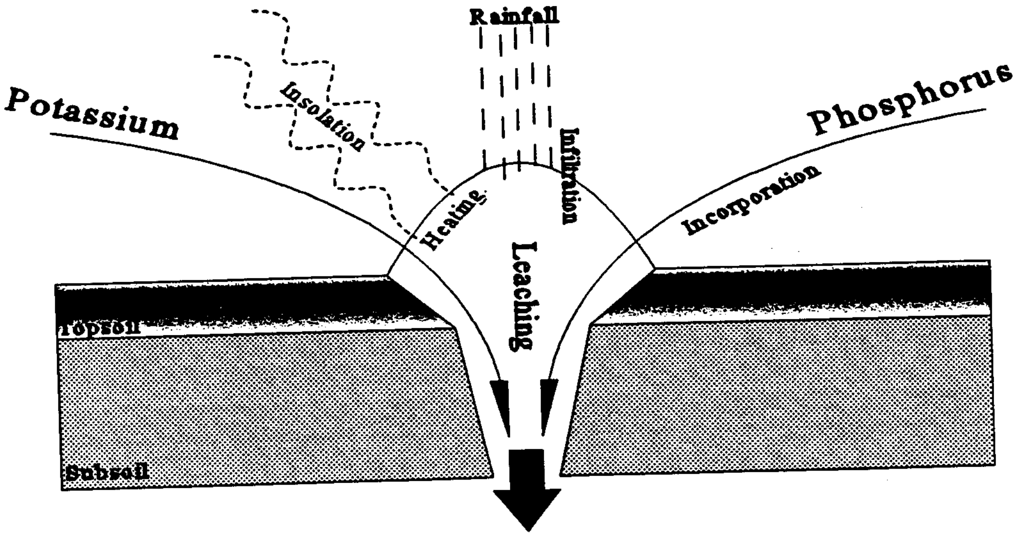

Figure 2. A depiction of an imported fire ant mound acting as a funnel, draining plant nutrients from soil landscapes.

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Neighborhood Fire Ant Abatement

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Queen relatedness in multiple-queen colonies of the fire ant *Solenopsis invicta* determined by microsatellite analysis

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Abstract: Genetic relatedness of queens within polygyne colonies of the red imported fire ant in central Texas was estimated by analyzing five microsatellite loci. The genotypes of 490 queens from 24 colonies revealed that relatedness among nestmate queens was not significantly different from zero. This finding confirms the observation of others that polygyne nests of *Solenopsis invicta* do not represent a society of closely related queens.

The Ants of East Texas: 25 years after

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In 1973, a survey (unpublished) was conducted to determine the ant species extant in eastern Texas. Four counties (Bexar, Cameron, Galveston and Nueces) were selected for the survey based upon their history to that point with regards to the imported fire ant, *Solenopsis invicta*. Although separated from the contiguously infested area of Texas until the mid 1980's (USDA Quarantine Maps), Bexar county has been under federal quarantine the longest of the four for red imported fire ants (1959). Galveston county was the next of the four to be declared officially infested by RIFA in 1969. Nueces county came under federal quarantine in 1973 (the initial year of the survey). Cameron county was the last to be placed under quarantine in 1996.

In 1997, the first of two follow-up surveys was undertaken to determine the change in RIFA and native ant status in east Texas.

Materials & Methods

Bait transects were run along major roadways within each county at 0.5 mile intervals. Care was made to attempt, whereever possible, to repeat the same lines of transect as used in the 1973 study. A liquid bait supplied by Dr. David Williams (USDA-ARS-CMAVE) was used in lieu of the sugar and meat baits used in the original survey. Bait traps were placed along roadsides and left for ca. one hour before retrieval. Within two hours of collection, specimens were removed from the traps; separated into distinct groupings and counted. Ants were placed in 70% ETOH and returned to the USDA facililty at Gulfport, MS where they were examined under magnification and identified to the lowest level possible. Dr. Daniel Wojcik (USDA-ARS-CMAVE) confirmed the identifications. Specimens are maintained at the Gulfport Plant Protection Station.

Results

In all four counties, increases in RIFA abundance (as a percentage of ants captured) were observed (Table 1). Both Bexar and Nueces counties had exceptionally large increases in RIFA numbers compared to the 1973 survey. These counties also reflected the largest decreases in ant species diversity (Table 2).The congenera of RIFA (*Solenopsis geminata* and *S. xyloni*) were completely eliminated in three of the four conties surveyed (Bexar, Galveston & Nueces). In 1973, *S. geminata* was found at 4.2% of the sites in Galveston county, 18.0% of the sites in Bexar county and 23.6% of the collection sites in Nueces county. Some decrease in *S. geminata* numbers was also noted in Cameron county. With *S. xyloni*, similar results were observed with decreases occurring in the three counties where it had been previously collected (Table 3).

Precipitous decreases in numbers of other ant species was noted in the

1997 survey. In Galveston county (1973), *Monomorium minimum* had been collected at 50.0% of the sites. By 1997, it could only be found at 4.8% of the collection points. *Tetramorium bicarinatum* dropped from 8.3% to 0.0%.

In Bexar county, *Pheidole dentata* had been collected at 32.5% of the sites in the first survey. In 1997, numbers had declined to 1.8%. During that same period, RIFA increased from zero collections to being collected at 36.9% of the sites. *Forelius pruinosus*, a species of ant that seems to compete well with RIFA (pers. obs.) also increased dramatically between the two surveys; going from 0.0% (1973) to 15.8% of the locations (1997).

Data from Nueces county reflected much the same changes as seen in Bexar county. RIFA numbers rose from 3.9% of the sites to 31.3%. Ten of the native ant species found in 1973 were not taken in the 1997 collections. One of these, *Pheidole floridana*, had previously been found at 7.9% of the locations.

In Cameron county, some decline in numbers of *S. geminata* was seen. A bigheaded ant, *Pheidole floridana*, was not taken in 1997. A congener, *Ph. dentata*, had also declined; falling from a high of 18.1% in 1973 to 3.5% in the 1997 sampling. *Monomorium minimum* likewise decreased sharply from its 1973 numbers (15.8 to 1.2%). Among other species, some increases had occurred. The harvester ant, *Pogonomyrmex barbatus*, was taken at 13.8% of the sites in 1997 as compared to 7.9% in 1973. A species of *Paratrechina*also increased; going from 0.8% to 6.9%. Six species of ants found in 1973 were not found in 1997. Conversely, seven new species were collected in 1997 that had not been taken in 1973. Two species not collected in 1973 were taken a a significant number of collection sites. An unidentified *Forelius* species in 1973 was captured at 9.8% of the sites in 1997. A yellow form of *Dorymyrmex* was taken at 4.6% of the sites in 1997. Another survey is scheduled for the spring of 1998 to eliminate any seasonal bias.

TABLE 1. Red Imported Fire Ants as a Percentage of Ants Collected.

COUNTY	1973	1997
Galveston	35.0	74.7
Nueces	8.9	90.4
Bexar	0.0	71.7
Cameron	0.0	6.4

TABLE 2. Changes in Diversity of Ant Species Captured in Four Texas Counties: 1973-1997.

	1973		1997	
COUNTY	# GEN.	# SPP.	# GEN.	# SPP.
Galveston	8	10	8	8
Nueces	7	13	5	5
Bexar	6	9	5	5
Cameron	10	13	10	13

Table 3. Species List of Ants Captured By Bait Trap in Four Texas Counties in 1973 and 1997.

GALVESTON CO.	NUMBER COLLECTED		MEAN NUMBER PER SITE		PERCENT OF SITES	
TAXON	1973	1997	1973	1997	1973	1997
Solenopsis invicta	452	700	37.6	38.9	**50.0**	**28.6**
Monomorium minimum	367	72	30.2	24.0	**50.0**	**4.8**
Paratrechina terricola	27	143	5.4	11.0	20.8	20.6
Tetramorium bicarinatum	151		75.5		**8.3**	**0.0**
Crematogaster sp.	161		161.0		**4.2**	**0.0**
S. geminata	120		120.0		**4.2**	**0.0**
Pheidole dentata	10	8	10.0	2.7	4.2	4.8
Forelius pruinosus	3	6	3.0	3.0	4.2	3.2
S. xyloni	1		1.0		**4.2**	**0.0**
Cardiocondyla nuda	1		1.0		**4.2**	**0.0**
Aphaenogaster texana		5		5.0	0.0	1.6
Crematogaster pilosus		2		2.0	0.0	1.6
Pseudomyrmex pallidus		1		1.0	0.0	1.6

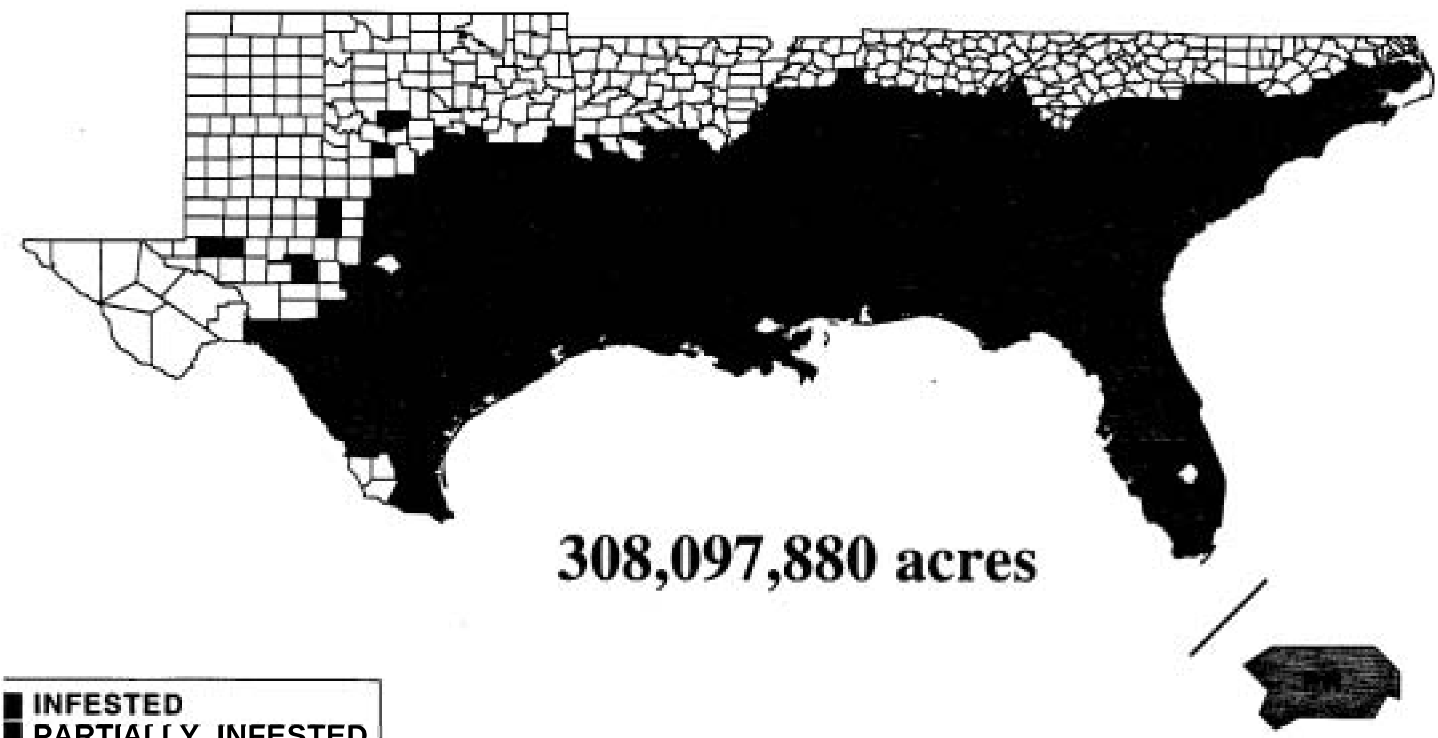
BEXAR CO.	NUMBER COLLECTED		MEAN NUMBER PER SITE		PERCENT OF SITES	
TAXON	1973	1997	1973	1997	1973	1997
Ph. dentata	455	76	12.0	76.0	**32.5**	**1.8**
S. geminata	471		22.4		**18.0**	**0.0**
M. minimum	140		28.0		**4.3**	**0.0**
Pheidole tepicana	39		9.8		**3.4**	**0.0**
S. xyloni	80		40.0		**1.7**	**0.0**
Pheidole floridana	12		12.0		0.9	0.0
P. terricola	9		9.0		0.9	0.0
Crematogaster laeviuscula	2		2.0		0.9	0.0
Dorymyrmex sp.	1		1.0		0.9	0.0
S. invicta		1046		49.8	**0.0**	**36.9**
F. pruinosus		250		27.8	**0.0**	**15.8**
Pogonomyrmex barbatus		31		15.5	**0.0**	**3.5**
Leptothorax nr schmitti		56		56.0	0.0	1.8

Table 3. Cont.

NUECES CO. TAXON	NUMBER COLLECTED 1973	NUMBER COLLECTED 1997	MEAN NUMBER PER SITE 1973	MEAN NUMBER PER SITE 1997	PERCENT OF SITES 1973	PERCENT OF SITES 1997
S. geminata	1658		55.3		**23.6**	**0.0**
S. xyloni	1769		63.2		**22.0**	**0.0**
Ph. dentata	621	3	51.8	3.0	**9.5**	**1.3**
Ph. floridana	416		41.6		**7.9**	**0.0**
S. invicta	449	1106	99.8	44.2	**3.9**	**31.3**
F. pruinosus	141	64	28.2	21.3	3.9	3.8
M. minimum	75		15.0		**3.9**	**0.0**
Ph. tepicana	356		89.0		**3.1**	**0.0**
S. (Diplorhoptrum) sp.	40		10.0		**3.1**	**0.0**
C. laeviuscula	27		13.5		1.6	0.0
C. nuda	2		1.0		1.6	0.0
Paratrechina sp.	2		2.0		0.8	0.0
P. terricola	3	46	3.0	4.6	**0.8**	**12.5**
Dorymyrmex sp.		4		1.3	**0.0**	**3.8**

CAMERON CO. TAXON	NUMBER COLLECTED 1973	NUMBER COLLECTED 1997	MEAN NUMBER PER SITE 1973	MEAN NUMBER PER SITE 1997	PERCENT OF SITES 1973	PERCENT OF SITES 1997
S. geminata	3817	1333	61.6	49.4	**48.8**	**31.0**
Ph. floridna	953		31.8		**23.6**	**0.0**
F. pruinosus	635	554	23.5	36.9	21.3	17.2
Ph. dentata	390	92	17.0	30.7	**18.1**	**3.5**
M. minimum	665	3	33.3	3.0	**15.8**	**1.2**
P. barbatus	57	85	5.7	7.1	**7.9**	**13.8**
C. laeviuscula	148	1	124.0	1.0	1.6	1.2
S. (Diplorhoptrum) sp.	2	40	2.0	40.0	0.8	1.2
Paratrechina sp.	1	64	1.0	10.7	**0.8**	**6.9**
P. terricola	1		1.0		0.8	0.0
Tetramorium similimum	1		1.0		0.8	0.0
Cardiocondyla emeryi	1		1.0		0.8	0.0
Macromischa subditiva	1		1.0		0.8	0.0
S. invicta		163		163.0	**0.0**	**1.2**
Forelius sp.		40		5.0	**0.0**	**9.8**
Dorymyrmex sp.		127		31.8	**0.0**	**4.6**
Pheidole sp.		2		2.0	0.0	1.2
Leptothorax nr. schmitti		64		64.0	0.0	1.2
A. texana		1		1.0	0.0	1.2

IMPORTED FIRE ANT QUARANTINE - 1998
Federal Register, January 28, 1998
7 CFR Part 301.81



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